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Crop Stress Detection and Classification Using Hyperspectral Remote Sensing

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CROP STRESS DETECTION AND CLASSIFICATION USING HYPERSPECTRAL REMOTE SENSING

By

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Agricultural production has observed many changes in technology over the last 20 years. Producers are able to utilize technologies such as site-specific applicators and remotely sensed data to assist with decision making for best management practices which can improve crop production and provide protection to the environment. It is known that plant stress can interfere with photosynthetic reactions within the plant and/or the physical structure of the plant. Common types of stress associated with agricultural crops include herbicide induced stress, nutrient stress, and drought stress from lack of water. Herbicide induced crop stress is not a new problem. However, with increased acreage being planting in varieties/hybrids that contain herbicide resistant traits, herbicide injury to non-target crops will continue to be problematic for producers. With rapid adoption of herbicide-tolerant cropping systems, it is likely that herbicide induced stress will continue to be a major concern. To date, commercially available herbicide-tolerant varieties/hybrids contain traits which allow herbicides like glyphosate and glufosinate-ammonium to be applied as a broadcast application during the growing season. Both glyphosate and glufosinate-ammonium are broad spectrum herbicides which have activity on a large number of plant species, including major crops like non-transgenic soybean, corn, and cotton. Therefore, it is possible for crop stress from herbicide applications to occur in neighboring fields that contain susceptible crop varieties/hybrids. Nutrient and moisture stress as well as stress caused by herbicide applications can interact to influence yields in agricultural fields. If remotely sensed data can be used to accurately identify specific levels of crop stress, it is possible that producers can use this information to better assist them in crop management to maximize yields and protect their investments. This research was conducted to evaluate classification of specific crop stresses utilizing hyperspectral remote sensing.

Key words: crop stress, herbicide drift, remote sensing

DEDICATION

The dedication of this research goes first and foremost to my Lord and Savior Jesus Christ. With Him, anything is possible. Next, I would like to dedicate this research to my parents, Tom and Jean Irby. Their love, support, teaching, encouragement, and understanding have made me the man that I am today. I would also like to dedicate this research to my sister, Tommi Odom and her family, Guy, Sy, and Ely Odom. Also, this research is dedicated to my brother, Ty Irby and his family, Ashlea, Taylor, Rylea, and Jon Isaac Irby. My siblings have meant much to me in my life and without them I certainly would not be where I am today. And last, but certainly not least, I would like to dedicate this research to my loving wife Megan. Her love, encouragement, and support have guided me throughout the majority of my education. Without her by my side, this journey would not have been possible. I love you all.

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I would also like to thank the other members of my committee, namely, Dr. Lori M. Bruce, Dr. Michael S. Cox, Dr. Jeffrey L. Willers, and Dr. Darrin F. Roberts, for the aid and direction they have shown me throughout this degree. I very much appreciate their willingness to work with me and help me achieve my goals to finish this degree.

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER I

INTRODUCTION

Agricultural production has gone through many changes in technology over the last 20 years. Producers are able to utilize technologies such as site-specific applicators and remotely sensed data to assist with improving crop production while providing protection to the environment. Tools which collect remotely sensed data, such as multispectral and hyperspectral sensors, can be utilized as a means to assess specific conditions within a given field which relate to crop yield (Seidl et al. 2004). For example, crop stress induced by pests, moisture or nutrient availability, or the crops reaction to a specific management practice can be monitored with remote sensing tools (Tartachnyk et al. 2006, Lichtenthaler 1996). Therefore, remote sensing systems have proven to be useful for many applications in production agriculture. These applications include detecting crop stress from lack of nutrients and moisture (Barnes et al. 2000), weed infestations (Chang et al. 2004), yield performance (Chang et al. 2003), crop stand density (Thorp et al. 2008), and injury from herbicide applications (Thelen et al. 2004, Henry et al. 2004). Ground-based systems, aerial imagery, and satellite imagery are options for obtaining remotely sensed data. Everman et al. (2008) utilized both a handheld spectroradiometer and aerial imagery to evaluate the effects of herbicides on the spectral reflectance of corn. Satellite imagery has been used to measure the nutrient uptake of winter cover crops (Hively et al. 2009). The use of such systems can provide valuable information about plant health from reflectance measurements of the crop canopy.

Herbicide induced stress on a crop is not a new problem. However, with increased acreage being planting in varieties/hybrids that contain herbicide resistant traits, herbicide injury to non-target crops will increasingly become problematic for producers. With rapid adoption of herbicide-tolerant cropping systems, it is likely that herbicide induced stress will continue to be a major concern. To date, commercially available herbicide-tolerant varieties/hybrids contain traits which allow herbicides like glyphosate and glufosinate-ammonium to be applied as a broadcast application during the growing season. Both glyphosate and glufosinate-ammonium are broad spectrum herbicides which have activity on a large number of plant species, including major crops like non-transgenic soybeans, corn, and cotton. Therefore, it is possible for crop stress from herbicide applications to occur in neighboring fields that contain susceptible crop varieties/hybrids. Crop stress from herbicides is often a result of unintentional applications. This phenomenon is often referred to as off-target herbicide deposition. The Environmental Protection Agency (EPA) defines spray drift as “The physical movement of a pesticide through air at the time of application or soon thereafter, to any site other than that intended for application (often referred to as off-target)” (Environmental Protection Agency 2009). There are many variables which can influence off-target deposition of herbicides. These factors include environmental conditions at time of application (i.e. wind speed, temperature, and humidity), herbicide formulation, application pressure, application speed, boom height, nozzle type, and droplet size (Carlsen et al. 2006).

A major challenge in an off-target herbicide incident is that injury may occur that cannot be detected by the human eye but can still cause yield reductions. Therefore, a producer may not realize that crop injury has occurred until harvest. Previous research has shown that ultra-low rates of glyphosate can reduce corn yield (Rowland 2000). Experiments conducted to simulate glyphosate drift in corn showed yield reductions of 78, 43, and 22% for simulated applications of 140, 70, and 35 grams acid equivalent per hectare (g ae/ha) of glyphosate, respectively (Ellis et al. 2003). Roider et al. (2007) found a 43% decrease in wheat yield when glyphosate was applied at 70 grams active ingredient per hectare (g ai/ha), which is approximately 6% of the normal use rate for this herbicide. Ellis et al. (2003) found height reductions and foliage discoloration from sub-lethal applications of glyphosate to both rice and corn crops were minimal, but negative effects on yields were significant.

Nitrogen and moisture stress as well as stress caused by herbicide applications can interact to influence yields in agricultural fields. Remotely sensed data can be used as a tool to assess these stresses (Barnes et al. 2000). The theory behind the utilization of remote sensing to detect plant stress is based on the assumption that stress is interfering with photosynthetic reactions within the plant or the physical structure of the plant and therefore affects the absorption of energy from light which changes the reflectance of energy from the plants (Riley et al. 1989, Hatfield and Pinter 1993). The nitrogen content found in leaves of many crops is an important indicator of growth status, quality, and yield (Cui et al. 2009). Nitrogen stress reduces the amount of chlorophyll and can result in increased reflectance of photosynthetically active light (Clay et al. 2006) and decreased reflectance in the near-infrared light (Cui et al. 2009, Yoder and Pettigrew-Crosby 1995). Water stress can influence reflectance due to reduced photochemical activity of chlorophyll (Clay et al. 2006, Souza et al. 2004). Herbicide applications to sensitive crops have also shown variations in reflectance. A reduction in NIR reflectance in corn was found when glyphosate was applied at 0.433 kg ae/ha, which is approximately 50% of the normal use rate for this herbicide (Irby 2009). Vegetation has unique characteristics regarding solar irradiance. Reflectance in the visible light spectrum (400-700 nm) is very low, transmittance is zero, and absorptance is high (Thelen et al. 2004). In the near-infrared (NIR) portion of the spectrum (700-1350 nm), both reflectance and transmittance are high and absorptance is low (Thelen et al. 2004). Because of these characteristics of reflectance of vegetation, multiple spectral vegetation indices have been developed. The normalized difference vegetation index (NDVI) is commonly used as an indication of plant vigor. NDVI is the ratio of NIR-Red/NIR+Red. Clay et al. (2006) used NDVI to measure water and nitrogen stress in corn while Henry et al. (2004) used NDVI to classify herbicide injury to soybeans and corn. Crop water stress indices (CWSI) have been used to map water stress in crops. This index uses canopy temperature and environmental conditions to calculate a value which describes water stress on a scale from 0 to

1, with 0 being no water stress and 1 being complete water stress. Barnes et al. (2000) used the CWSI paired with NDVI to map water stress in cotton. Experiments have shown that hyperspectral vegetation indices can provide a more accurate assessment of crop parameters than did equivalent indices from multispectral sensors (Thenkabail et al. 2002). However, hyperspectral data are more complicated when compared to multispectral data due to the volume of data which is obtained (Karimi et al. 2005). For example, data collected with the SpecTIR™ aerial hyperspectral imager contain 128 bands ranging from 400 to 994 nm with a spectral resolution of 3 nm at 700 nm and 10 nm at 1400/2100 nm (Anonymous 2012). In order to extract useful information from larger hyperspectral data sets, it is important to first select a range of wavelengths that can be used to describe information about the specific target.

Remotely sensed data can provide valuable information about the overall health of a plant in instances where crop injury is suspected but not visible. Some factors that should be considered when using remotely sensed data in this capacity include time of day/year, topography, soil type, and crop type. Previous research has shown that reflectance values in tilled fields were primarily influenced by soil characteristics during the early stages of the growing season (Huete et al. 1985). Chang et al. (2003) reported that characteristics of spectral reflectance are influenced by plant factors more than soil factors as the growing season progresses.

The ability to rapidly detect and assess herbicide induced stress to a crop would be beneficial in many aspects. From a producer’s standpoint, a rapid response time is needed in order to make a management decision about the stressed crop. In the event of herbicide injury to a crop, producers could use the information obtained from the remotely sensed data coupled with data showing yield reductions correlated to reflectance measurements to make informed decisions for replanting or leaving the injured crop in the field.

Remotely sensed data could also allow for a rapid detection tool in the event of agriterrorism, where intentional application of herbicides or biological agents was made in order to harm the nation’s food supply. Many commonly used herbicides are readily available and, if used intentionally to destroy crops, could have detrimental effects to our nation’s food supply. Utilizing remote sensing tools could allow assessment of the level and quantity of damage or the lack thereof of herbicides or biological agents intentionally applied to our food supply.

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CHAPTER II

THE DEVELOPMENT OF A GROUND-BASED SPECTRAL ACQUISITION SYSTEM TO EVALUATE OFF-TARGET HERBICIDE DEPOSITION

Introduction

Agricultural production systems have observed many changes in recent years. Technological advances have allowed producers to utilize tools such as internet, mobile phones, global positioning systems, site-specific applicators, and remotely sensed data to assist with decision making pertaining to best management practices to improve crop production and provide protection to the environment. Producers who are adopting these technologies can combine many of these tools to assess equipment performance, monitor environmental conditions, and evaluate the condition of their crops at any given time.

Tools which collect remotely sensed data, such as multispectral and hyperspectral sensors, can be utilized as a means to assess specific conditions within a given field which relate to crop yield (Seidl et al. 2004). For example, crop stress induced by pests, moisture or nutrient availability, or the crops reaction to a specific management practice can be monitored with remote sensing tools (Tartachnyk et al. 2006, Lichtenthaler 1996). Therefore, remote sensing systems have proven to be useful for many applications in production agriculture. These applications include detecting crop stress from lack of nutrients and moisture (Barnes et al. 2000), weed infestations (Chang et al. 2004), yield performance (Chang et al. 2003), crop stand density (Thorp et al. 2008), and injury from herbicide applications (Thelen et al. 2004, Henry et al. 2004). Ground-based systems, aerial imagery, and satellite imagery are options for obtaining remotely sensed data. Everman et al. (2008) utilized both a handheld spectroradiometer and aerial imagery to evaluate the effects of herbicides on the spectral reflectance of corn. Satellite imagery has been used to measure the nutrient uptake of winter cover crops (Hively et al. 2009). The use of such systems can provide valuable information about plant health from reflectance measurements of the crop canopy.

A particular area of interest where remotely sensed data can be a benefit is the evaluation of stress in a producer's crop. Examples of stress which can affect a crop and ultimately the crop's yield include nutrient stress, moisture stress, and herbicide stress. Herbicide induced stress on a crop is not a new problem. However, with increased acreage being planting in varieties/hybrids that contain herbicide resistant traits, herbicide injury to non-target crops continues to cause problems for producers. As the industry continues to develop new genetic traits that will allow multiple herbicide chemistries to be applied safely to the target crop, it is likely that herbicide induced stress will continue to be a major concern in non-target fields. To date, commercially available herbicide-tolerant varieties/hybrids contain traits which allow herbicides like glyphosate and glufosinate-ammonium to be applied as a broadcast application during the growing season. Both glyphosate and glufosinate-ammonium are broad spectrum herbicides which have activity on a large number of plant species, including major crops like non-transgenic soybeans, corn, and cotton. Therefore, it is possible for crop stress from herbicide applications to occur in neighboring fields that contain susceptible crop varieties/hybrids.

Crop stress from herbicides is often a result of unintentional applications. This phenomenon is often referred to as off-target herbicide deposition. The Environmental Protection Agency (EPA) defines spray drift as “The physical movement of a pesticide through air at the time of application or soon thereafter, to any site other than that intended for application (often referred to as off-target)” (Environmental Protection Agency 2009). There are many variables which can influence off-target deposition of herbicides. These factors include environmental conditions at time of application (i.e. wind speed, temperature, and humidity), herbicide formulation, application pressure, application speed, boom height, nozzle type, and droplet size (Carlsen et al. 2006).

A major challenge in an off-target herbicide incident is that injury may occur that cannot be detected by the human eye but can still cause yield reductions. Therefore, a producer may not realize that crop injury has occurred until harvest. Previous research has shown that ultra-low rates of glyphosate can reduce corn yield (Rowland 2000). Experiments conducted to simulate glyphosate drift in corn showed yield reductions of 78, 43, and 22% for simulated applications of 140, 70, and 35 grams acid equivalent per hectare (g ae/ha) of glyphosate, respectively (Ellis et al. 2003). Roider et al. (2007) found a 43% decrease in wheat yield when glyphosate was applied at 70 grams active ingredient per hectare (g ai/ha), which is approximately 6% of the normal use rate for this herbicide. Ellis et al. (2003) found that height reductions and foliage discoloration from sub-lethal applications of glyphosate to both rice and corn crops were minimal, but negative effects on yields were significant.

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Remotely sensed data can provide valuable information about the overall health of a plant in instances where crop injury is suspected but not visible. Some factors that should be considered when using remotely sensed data in this capacity include time of day/year, topography, soil type, and crop type. Previous research has shown that reflectance values in tilled fields were primarily influenced by soil characteristics during the early stages of the growing season (Huete et al. 1985). Chang et al. (2003) reported that characteristics of spectral reflectance are influenced by plant factors more than soil factors as the growing season progresses. This is expected due to the methods by which many of our agricultural crops are grown. Typically, row crops are planted in a variety of spacings both between the rows as well as within each row. These spacings are chosen based on the optimum plant population which will maximize yield and provide canopy cover to efficiently use light energy in the plant’s photosynthetic processes. Therefore, as the season progress, the crop canopy begins to cover the soil surface allowing spectral reflectance to be influenced more by plant factors rather than soil factors.

The ability to rapidly detect and assess herbicide induced stress to a crop would be beneficial in many aspects. From a producer’s standpoint, a rapid response time is needed in order to make a management decision about the stressed crop. In the event of herbicide injury to a crop, producers could use the information obtained from the remotely sensed data coupled with data showing yield reductions correlated to reflectance measurements to make informed decisions for replanting or leaving the injured crop in the field. This type of situation is often the most difficult decision that a producer might have to make. Factors such as the type of injury and time of year play important roles in this process. Having tools in place which can assist in rapidly making these decisions could allow producers to salvage their investment rather than accepting a total loss. Experiments have shown that hyperspectral data can provide a more accurate assessment of crop parameters than did information from multispectral sensors (Thenkabail et al. 2002). However, hyperspectral data are more complicated when compared to multispectral data (Karimi et al. 2005). One available tool to gather hyperspectral information about a crop is aerial imagery. Utilizing aerial imagery to gather spectral data is an excellent method for collecting information over a large area in a short amount of time. However, some forms of aerial imagery can be quite expensive. For example, a single hyperspectral image can provide a tremendous amount of information but can cost between $30,000 and $40,000. This is not a practical means for gathering spectral information about suspected stress in a crop. In addition, handheld sensors can often be tedious and may require more time to gather the same amount of information when compared to the aerial imagery. Therefore, it is important to develop an economic and efficient method for collecting this information to assist producers in making final decisions about handling stressed crops. The objective of this research was to develop a ground-based spectral acquisition system to be utilized for evaluation of off-target herbicide deposition.

Materials and Methods

Experiments were conducted at the Black Belt Branch Experiment Station in Brooksville, MS to compare classification results of hyperspectral data acquired by an aerial platform to data acquired through a ground-based spectral acquisition system. Field corn (*Zea mays*) was planted in a field measuring 2.3 hectares in size. The field was planted according to standard agricultural fertility and crop row spacing practices. The seeding rate for corn was 69,000 seeds per hectare. In order to gather as much spatial variability as possible, field plot size measured 7.70 meters wide by 30.5 meters long. Herbicide applications consisted of glufosinate-ammonium, the active ingredient in the herbicide Liberty® 280 SL. Glufosinate-ammonium was applied to a corn hybrid which is sensitive to this herbicide. Herbicide applications were made when corn reached the V6-V7 growth stage. Glufosinate-ammonium application rates included the recommended labeled rate (1X) of 0.59 kilograms of active ingredient per hectare (kg ai/ha) as well as 0.30, 0.15, 0.07, 0.04, and 0.02 kg ai/ha, which correspond to 1/2X, 1/4X, 1/8X, 1/16X, and 1/32X fractions of the recommended labeled rate. An untreated check was included for comparison purposes. Herbicide applications were made using a tractor mounted spray boom equipped with shields to minimize contamination to neighboring plots. All herbicides were applied at an application volume of 140 liters per hectare. Data collection consisted of hyperspectral data collected using the Analytical Spectral Devices (ASD™) Fieldspec Pro handheld spectroradiometer and the SpecTIR™ airborne hyperspectral imager. Ground-based hyperspectral data were collected over a 14 day period with collection timings of 1, 4, 7, and 14 days after herbicide application, depending on the weather. Due to the cost of the aerial hypserspectral imagery, a single image was collected 4 days after herbicide application. Handheld spectroradiometer data were collected in conjunction with a Topcon HiPer Lite Plus real time kinematic (RTK) global positioning system (GPS) to ensure that each data point received a fixed spatial information description. Principal component analysis and stepwise linear discriminant analysis techniques were utilized for selecting spectral features which can be utilized to describe the actual herbicide concentrations applied in the field. The resulting data were then utilized to generate classification matrixes providing classification accuracies of the system's capability for identifying spectral features associated with the various herbicide concentrations.

Results and Discussion

Remotely sensed data acquisition can be time consuming and difficult as well as expensive. This experiment was designed to develop a technique which utilizes the handheld ASD instrument and RTK GPS system together to gather spectral and spatial information from the stressed crop. In order to develop this technique, it was necessary to develop a method for integrating the ASD™ Fieldspec Pro handheld spectroradiometer and Topcon HiPer Lite Plus RTK GPS for on-the-go data collection (Figure 2.1).

Figure 2.1 Topcon HiPer Lite Plus RTK GPS and Fieldspec Pro handheld spectroradiometer integration.

The handheld spectroradiometer was set up in conjunction with the RTK GPS system with settings applied to assign a fixed latitude and longitude value with each recorded spectral record. The next step in the process was to secure a method for on-the-go data collection. A platform equipped with seating available for the operator(s) of the handheld spectroradiometer was mounted to the 3 point hitch of a tractor with the necessary ground clearance to move through the field without direct contact with the crop (Figure 2.2).

Figure 2.2 Tractor mounted platform for on-the-go data collection.

By applying a collection setting of the average of 10 spectral signature readings per second, it was possible to modify the speed of the tractor to achieve a specific sequence for collection of hyperspectral data as the machine moved through the field. For example, setting the machine speed to 4.8 kilometers per hour allowed the system to collect the average of 10 spectral signature readings every 1.3 meters as the machine moved through the field. This system offered the capability of collecting real time spectral information tagged with spatial locations automatically as a machine travels continuously through a field. Figure 2.3 illustrates only 1/10th of the actual locations where hyperspectral data sampling occurred. This system offered the capability of gathering a large volume of information from the target field in an efficient manner. For example, the experimental field measuring 2.3 ha in size required approximately 37 minutes to completely sample the field in 8 row increments. This provided 230 actual signature readings from each plot.

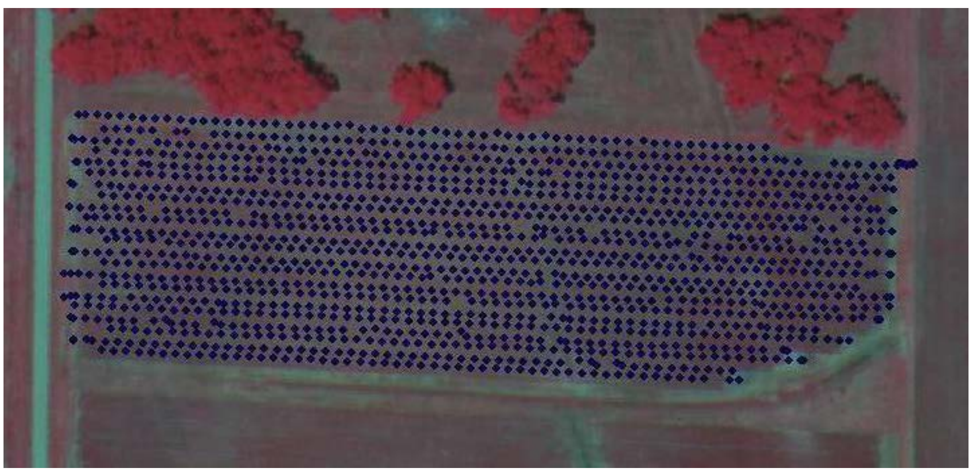

Figure 2.3 Map display of 1/10th of the total hyperspectral data sampling points collected during 1 sample timing.

These data were analyzed to provide assessments of the system's capability to accurately classify the applied herbicide concentration via measured spectral reflectance. In order to compare classification accuracies of the ground-based spectral acquisition system to the accuracies obtained from the aerial platform, only the information obtained 4 days after application was utilized for this experiment due to the fact that this was the only hyperspectral image obtained. Tables 2.1 to 2.4 each display a classification matrix for the classification accuracies that were obtained 4 days after treatment (DAT) when glufosinate-ammonium was applied at various rates to susceptible corn to simulate off-target herbicide deposition. These classification matrixes were generated utilized the results of principal component analysis (PCA) and stepwise linear discriminant analysis (SLDA) techniques which were applied to both the data collected with the ground-based spectral acquisition system and the aerial platform. Overall, producer, and consumer's accuracies were calculated for each classification matrix. The overall accuracy is the percentage of the correctly classified spectral features obtained with the specific system. Producer's accuracy is a measure of the system's capability to correctly classify spectral features which correspond to a specific herbicide concentration. In other words, the producer's accuracy is the percentage of spectral features that were classified with the correct herbicide concentration, while the remaining spectral features in this category which actually belonged with the correct concentration were classified as a different concentration. Consumer's accuracy is a measure of the spectral features that were correctly classified to correspond with the actual herbicide concentration that was applied. For example, herbicide concentration Z with a producer's accuracy of Y% and a consumer's accuracy of X% simply means that the system identified Y% of the spectral features as herbicide concentration Z, but only X% of the spectral features actually belonged with herbicide concentration Z.

The classification matrix resulting from the data generated by the PCA technique (Table 2.1) applied to the hyperspectral data acquired through the ground-based spectral acquisition system displays an overall accuracy of 24%. Producer's accuracy results were poor for this system, with the exception of the producer's accuracy for correctly identifying the 1/8X rate which received an accuracy of 80%. The consumer's accuracies were low for all rates ranging from 11 to 35%. The greatest overall accuracy of the data collected with the ground-based acquisition system of 69% was achieved with the SLDA system (Table 2.2). Producer's accuracies ranged from 54 to 84% while consumer's accuracies ranged from 59 to 84%. The system identified 75% of the spectral features as the untreated while 72% of the spectral features were actually associated with the untreated. Similarly, the producer's accuracy of 72% and consumer's accuracy of 68% for the 1/8X concentration indicates that the system identified 72% of the spectral features as the 1/8X concentration while 68% of the spectral features did indeed correspond with this concentration.

The classification matrixes generated from the data from the two analysis techniques applied to the SpecTIR hyperspectral aerial imagery are displayed in Tables 2.3 and 2.4. Similar results were observed for the data collected with the aerial platform (Table 2.3) compared to that of the ground-based acquisition system and subjected to the PCA technique (Table 2.1). An overall accuracy of 17% was observed with producer's accuracies ranging from 0 to 44% (Table 2.3). The classification matrixes generated from the data subjected to the PCA technique consistently resulted in low producer's accuracies for the three lowest application rates. Consumer's accuracies were not achieved for these rates. The consumer's accuracies observed for the 1/4X, 1/2X, and 1X rates were 20, 16, and 15%, respectively. As was the case with the data acquired with the ground-based system, a higher overall accuracy was observed when the SLDA technique was applied to the aerial imagery data resulting in an overall accuracy of 77% (Table 2.4). Producer's accuracies for the untreated, 1/8X, 1/4X, 1/2X, and 1X concentrations were 91, 77, 91, 84, and 88%, respectively. The producer's accuracies observed for the two lowest concentrations of 1/16X and 1/32X were 49 and 52%. Consumer's accuracies ranged from 59 to 88% for all treatments.

Conclusion

While it is often unknown what herbicide actually was involved in cases of off-target deposition, it is likely that the spectral response will be influenced within a matter of days after the occurrence due to the fact that some herbicides begin to influence plant structure in this amount of time. For example, plants exposed to a labeled rate of glufosinate-ammonium will exhibit necrosis of leaves and young shoots within 2 to 4 days after the application if conditions are favorable (Anonymous 2012). This is increasingly important for the producers whose crop was affected so that management decisions can be made regarding the injured crop. The results from these experiments indicate that the ground-based spectral acquisition system can be utilized to collect spectral information which provides useful insights to the level of injury to the crop after an off-target herbicide deposition occurrence within 4 days after the incident. Similar results were observed for both the ground-based system and the more expensive aerial platform system. These data also demonstrate that the ground-based system had higher producer’s accuracies for the lower herbicide concentrations of 1/16X and 1/32X of glufosinate-ammonium when compared to the results from the aerial imagery subjected to the SLDA technique. This information could prove useful in the event of off-target herbicide deposition as it is likely that lower herbicide concentrations will be difficult to observe visually. Producer’s accuracies observed with the data from the ground-based spectral acquisition system for these two concentrations were 67% with consumer accuracies of 59 and 71% for the 1/16X and 1/32X concentrations, respectively. In other words, this system identified 67% of the spectral features associated with the 1/16X and 1/32X concentrations while 59 and 71% of the observed spectral features actually belonged to the two concentrations. These data indicate that the system could prove valuable for classifying spectral features associated with these low concentrations. If a ground-based system such as this can be implemented in a timely manner, it is possible to provide the producer with an estimate of damage that the crop may or may not have received. This could allow for proper decisions to be made regarding the overall health of the current crop and whether or not the level of injury presents a financial liability should the crop be left as is.

Table 2.1 Classification matrix resulting from data generated through principal component analysis of hyperspectral data obtained through a ground-based spectral acquisition system 4 days after treatment of various rates of glufosinate-ammonium applied to susceptible corn.

		Herbicide Concentration[1]							
		Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	1X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	0	0	5	23	5	24	30	0%
	1/32X	0	0	1	32	4	17	26	0%
	1/16X	0	0	8	12	1	18	35	11%
	1/8X	0	0	2	65	2	1	11	80%
	1/4X	0	0	10	26	3	14	38	3%
	1/2X	0	0	5	15	4	25	43	27%
	1X	0	0	7	12	8	23	42	46%
	Consumer's Accuracy[3]	--	--	21%	35%	11%	21%	19%	24%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.59 kg ai/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

Table 2.2 Classification matrix resulting from data generated through stepwise linear discriminant analysis of hyperspectral data obtained through a ground-based spectral acquisition system 4 days after treatment of various rates of glufosinate-ammonium applied to susceptible corn.

		Herbicide Concentration[1]							
		Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	1X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	65	15	5	0	1	1	0	75%
	1/32X	15	54	9	0	2	0	1	67%
	1/16X	8	8	50	3	2	4	0	67%
	1/8X	1	9	4	59	4	5	0	72%
	1/4X	0	0	4	8	56	13	8	63%
	1/2X	1	0	1	0	16	50	24	54%
	1X	0	1	1	0	1	12	79	84%
	Consumer's Accuracy[3]	72%	71%	59%	68%	84%	68%	62%	69%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.59 kg ai/ha.

[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.

[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.

[4] – Overall accuracy of the system.

Table 2.3 Classification matrix resulting from data generated through principal component analysis of hyperspectral data obtained through SpecTIR imagery 4 days after treatment of various rates of glufosinate-ammonium applied to susceptible corn.

		Herbicide Concentration[1]							
		Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	1X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	9	0	0	0	45	162	164	2%
	1/32X	6	0	0	0	57	120	148	0%
	1/16X	6	0	0	0	48	127	140	0%
	1/8X	5	0	0	0	60	149	172	0%
	1/4X	3	0	0	0	77	124	164	21%
	1/2X	1	0	0	0	51	165	189	41%
	1X	6	0	0	0	56	160	176	44%
	Consumer's Accuracy[3]	25%	--	--	--	20%	16%	15%	17%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.59 kg ai/ha.

[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.

[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.

[4] – Overall accuracy of the system.

Table 2.4 Classification matrix resulting from data generated through stepwise linear discriminant analysis of hyperspectral data obtained through SpecTIR imagery 4 days after treatment of various rates of glufosinate-ammonium applied to susceptible corn.

		Herbicide Concentration[1]							
		Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	1X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	344	22	8	5	1	0	0	91%
	1/32X	122	171	27	9	2	0	0	52%
	1/16X	77	58	156	26	4	0	0	49%
	1/8X	44	7	18	296	21	0	0	77%
	1/4X	0	0	4	6	335	16	7	91%
	1/2X	0	0	0	0	17	341	48	84%
	1X	0	0	0	0	0	49	349	88%
	Consumer's Accuracy[3]	59%	66%	73%	87%	88%	84%	86%	77%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.59 kg ai/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

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CHAPTER III

EVALUATION OF CROP SPECTRAL FEATURES CONTAINING SPATIAL INFORMATION COLLECTED AFTER HERBICIDE INDUCED STRESS

Introduction

In recent years, producers have observed an increase in observations of crop stress due to herbicide drift. Herbicide drift has been referred to as an off-target movement that could cause serious injury if contact is made with susceptible plants (Deeds et al. 2006). This phenomenon of herbicide induced stress on crops is not new. Typically, stress caused by herbicides occurs during an off-target deposition instance of a herbicide application in the proximity of a field where susceptible plants are growing. The Environmental Protection Agency (EPA) defines spray drift as “The physical movement of a pesticide through air at the time of application or soon thereafter, to any site other than that intended for application (often referred to as off-target)” (Environmental Protection Agency 2009). There are many variables which can influence off-target deposition. These factors include environmental conditions at time of application (i.e. wind speed, temperature, and humidity), herbicide formulation, application pressure, application speed, boom height, nozzle type, and droplet size (Carlsen et al. 2006).

Problems associated with off-target herbicide deposition have been around for quite some time. Ellis et al. (2003) proposed that an expected increase in transgenic cropping systems would increase the use of glyphosate causing an increase in potential for off-target herbicide movement. With increased acreage being planting in varieties/hybrids that contain herbicide resistant traits, herbicide injury to non-target crops may continue to cause problems for producers. As the industry continues to develop new genetic traits that will allow multiple herbicide chemistries to be applied safely to the target crop, it is likely that herbicide induced stress will continue to be a major concern in non-target fields. To date, commercially available herbicide-tolerant varieties/hybrids contain traits which allow herbicides like glyphosate and glufosinate-ammonium to be applied as a broadcast application during the growing season. Both glyphosate and glufosinate-ammonium are broad spectrum herbicides which have activity on a large number of plant species, including major crops like non-transgenic soybeans, corn, cotton, rice, and wheat. Both of these herbicides are commonly used to control existing weeds in fields prior to planting. Typically, cotton and soybean crops are planted later in the season than rice and corn. Therefore, spring burndown applications as well as early postemergence applications can be opportunities for off-target deposition where established susceptible crops are growing within close proximities of fields containing transgenic crops.

One of the major challenges associated with off-target herbicide deposition is having the capability to assess the level of crop injury or the lack thereof. It is possible that injury may occur that cannot be detected by the human eye but can still cause yield reductions. Therefore, a producer may not realize that crop injury has occurred until harvest. Previous research has shown that off-target deposition cases often involve herbicide concentrations between 1/100 and 1/10 of the labeled herbicide rates (Al-Khatib et al. 2003, Al-Khatib and Peterson 1999, Al-Khatib and Tamhane 1999, Al-Khatib et al. 1993, Bode 1987, Maybank et al. 1978). Corn injury has been observed for glyphosate drift rates as low as 11 g/ha (Al-Khatib et al. 2003, Al-Khatib et al. 2000). Other experiments conducted to simulate glyphosate drift in corn showed yield reductions of 78, 43, and 22% for simulated applications of 140, 70, and 35 grams acid equivalent per hectare (g ae/ha) of glyphosate, respectively (Ellis et al. 2003). Roider et al. (2007) found a 43% decrease in wheat yield when glyphosate was applied at 70 grams active ingredient per hectare (g ai/ha), which is approximately 6% of the normal use rate for this herbicide. Ellis et al. (2003) found that height reductions and foliage discoloration from sub-lethal applications of glyphosate to both rice and corn crops were minimal, but negative effects on yields were significant.

Technological advances have changed agricultural production systems. One example of this change is the use of remotely sensed data to monitor the condition of a crop at a given time and location. Currently available tools which can be utilized to monitor crop stress include multispectral and hyperspectral sensors. These tools can be utilized as a means to assess specific conditions within a given field which relate to crop yield (Seidl et al. 2004). For example, crop stress induced by pests, moisture or nutrient availability, or the crops reaction to a specific management practice can be monitored with remote sensing tools (Tartachnyk et al. 2006, Lichtenthaler 1996). Therefore, remote sensing systems have proven to be useful for many applications in production agriculture. Remote sensing systems have been utilized for detecting crop stress from lack of nutrients and moisture (Barnes et al. 2000), weed infestations (Chang et al. 2004), yield performance (Chang et al. 2003), crop stand density (Thorp et al. 2008), and injury from herbicide applications (Thelen et al. 2004, Henry et al. 2004). Ground-based systems, aerial imagery, and satellite imagery are options for obtaining remotely sensed data. Everman et al. (2008) utilized both a ground-based handheld spectroradiometer and aerial imagery to evaluate the effects of herbicides on the spectral reflectance of corn. Satellite imagery has been used to measure the nutrient uptake of winter cover crops (Hively et al. 2009). The use of such systems can provide valuable information about plant health through reflectance measurements of the crop canopy.

The theory behind the utilization of remote sensing to detect plant stress is based on the assumption that stress is interfering with photosynthetic reactions within the plant or the physical structure of the plant and therefore affects the absorption of energy from light which changes the reflectance of energy from the plants (Riley et al. 1989, Hatfield and Pinter 1993). Herbicide applications to sensitive crops have shown variations in reflectance. A reduction in NIR reflectance in corn was found when glyphosate was applied at 0.433 kg ae/ha, which is approximately 50% of the normal use rate for this herbicide (Irby 2009). Vegetation has unique characteristics regarding solar irradiance. Reflectance in the visible light spectrum (400-700 nm) is very low, transmittance is zero, and absorptance is high (Thelen et al. 2004). In the near-infrared (NIR) portion of the spectrum (700-1350 nm), both reflectance and transmittance are high and absorptance is low (Thelen et al. 2004). Because of these characteristics of reflectance of vegetation, multiple spectral vegetation indices have been developed. The normalized difference vegetation index (NDVI) is commonly used as an indication of plant vigor. NDVI is the ratio of NIR-Red/NIR+Red. Clay et al. (2006) used NDVI to measure water and nitrogen stress in corn while Henry et al. (2004) used NDVI to classify herbicide injury to soybeans and corn. A common method for collection of multispectral data is through aerial mounted platforms. This method is relatively inexpensive and can provide useful information about a crop at the field level. Experiments have shown, however, that hyperspectral data can provide a more accurate assessment of crop parameters than did equivalent data from multispectral sensors (Thenkabail et al. 2002).

Remotely sensed data can provide valuable information about the overall health of a plant in instances where crop injury is suspected but not visible. Some factors that should be considered when using remotely sensed data in this capacity include time of day/year, topography, soil type, and crop type. Previous research has shown that reflectance values in tilled fields were primarily influenced by soil characteristics during the early stages of the growing season (Huete et al. 1985). Chang et al. (2003) reported that characteristics of spectral reflectance are influenced by plant factors more than soil factors as the growing season progresses. This is expected due to the methods by which many of our agricultural crops are grown. Typically, row crops are planted in a variety of spacings both between the rows as well as within each row. These spacings are chosen based on the optimum plant population which will maximize yield and provide canopy cover to efficiently use light energy in the plant's photosynthetic processes. Therefore, as the season progress, the crop canopy begins to cover the soil surface allowing spectral reflectance to be influenced more by plant factors rather than soil factors.

The ability to rapidly detect and assess herbicide induced stress to a crop would be beneficial in many aspects. From a producer's standpoint, a rapid response time is needed in order to make a management decision about the stressed crop. In the event of herbicide injury to a crop, producers could use the information obtained from the remotely sensed data coupled with data showing yield reductions correlated to reflectance measurements to make informed decisions for replanting or leaving the injured crop in the field. This type of situation is often the most difficult decision that a producer might have to make. Factors such as the type of injury and time of year play important roles in this process. Having tools in place which can assist in rapidly making these decisions could allow producers to salvage their investment rather than accepting a total loss. Therefore, it is important to evaluate remotely sensed information for classifying spectral features associated with specific sub-lethal concentrations of a herbicide in the event of off-target herbicide deposition. The objective of this research was to evaluate spectral features associated with various sub-lethal concentrations of herbicides applied to susceptible crops.

Materials and Methods

Experiments were conducted at the Black Belt Branch Experiment Station in Brooksville, MS to evaluate spectral features associated with various concentrations of herbicides applied to susceptible crops. Field corn (*Zea mays*) was planted in 2 experimental fields which are 2.3 and 3.2 hectares in size. Wheat (*Triticum aestivium*) was planted in a single experimental field which was 3.2 hectares in size. Each field was planted according to standard agricultural practices for each crop. The seeding rate for corn and wheat was 69,000 seeds per hectare and 95 kilograms per hectare, respectively. In order to gather as much spatial variability as possible, field plot size measured 7.70 meters wide by 30.5 meters long. Herbicide applications included glufosinate-ammonium, the active ingredient in the herbicide Liberty® 280 SL, clethodim, the active ingredient in the herbicide Select Max®, and glyphosate, the active ingredient in the herbicide Roundup®. Glufosinate-ammonium and clethodim were applied to a corn hybrid which is sensitive to both of these herbicides. Glyphosate was applied to wheat which is highly sensitive to this herbicide. Herbicide applications were made when corn reached the V6-V7 growth stage and when wheat reached the boot stage. Glufosinate-ammonium application rates included the recommended labeled rate (1X) of 0.59 kilograms of active ingredient per hectare (kg ai/ha) as well as 0.30, 0.15, 0.07, 0.04, and 0.02 kg ai/ha, which correspond to 1/2X, 1/4X, 1/8X, 1/16X, and 1/32X fractions of the recommended labeled rate.

Similarly, clethodim rates were based on the recommended labeled rate of 0.10 kg ai/ha and included concentrations of 0.05, 0.025, 0.013, 0.006, and 0.003 kg ai/ha, which correspond to 1/2X, 1/4X, 1/8X, 1/16X, and 1/32X fractions of the recommended labeled rate. Glyphosate rates for the applications to wheat were based on the recommended labeled rate of 0.86 kilograms of acid equivalent per hectare (kg ae/ha) and included concentrations of 0.43, 0.11, and 0.03 kg ae/ha which correspond to 1/2X, 1/8X, and 1/32X fractions of the recommended labeled rate. Data collection consisted of hyperspectral data collected using the Analytical Spectral Devices (ASD™) Fieldspec Pro handheld spectroradiometer. Hyperspectral data were collected over a 14 day period for each crop with collection timings of 1, 4, 7, and 14 days after herbicide application, depending on the weather. Handheld spectroradiometer data were collected in conjunction with a Topcon HiPer Lite Plus real time kinematic (RTK) global positioning system (GPS) to ensure that each data point received a fixed spatial information description. Visual injury ratings were recorded 7, 14, and 28 days after application and were based on a scale of 0 to 100, where 0 = no crop injury and 100 = complete crop death (Frans et al. 1986). At harvest, machine harvested yields were collected from the two center rows of each corn plot and the center 1.8 meters of each wheat plot. Principal component analysis (PCA), linear discriminant analysis (LDA), stepwise linear discriminant analysis (SLDA), a multi-classifier decision fusion (MCDF), and a discrete wavelet transfer multi-classifier decision fusion (DWT-MCDF) analysis techniques were tested in order to confirm which technique provided consistent results. Principal component analysis, LDA, and SLDA techniques are commonly used for dimensionality reduction as well as feature extraction in hyperspectral data (Kalluri et al. 2009). The MCDF technique separates the hyperspectral data into multiple subsets allowing classification of each subset and ultimately a single identification of each class per hyperspectral signature (Prasad and Bruce 2008). Ultimately, data were analyzed using a MCDF technique and the results of this technique were utilized to generate classification accuracies of the hyperspectral data acquired with a ground-based spectral acquisition system after application of the various herbicide rates applied in the field.

Results and Discussion

Hyperspectral data acquired through a ground-based spectral acquisition system were analyzed to provide assessments of the system's capability to accurately predict the herbicide rate that was actually applied based on the collected spectral information. Typically, herbicides will influence the plant structure within a matter of days after direct contact. Therefore, these data were collected multiple times over a 14 day period after simulation of off-target herbicide deposition. Tables 3.1 to 3.11 display classification matrixes of the handheld hyperspectral data. Overall, producer, and consumer's accuracies were calculated for each classification matrix. The overall accuracy is the percentage of the correctly classified spectral features obtained with the specific system. Producer's accuracy is a measure of the system's capability to correctly classify spectral features which correspond to a specific herbicide concentration. In other words, the producer's accuracy is the percentage of spectral features that were classified with the correct herbicide concentration, while the remaining spectral features in this category which actually belonged with the correct concentration were classified as a different concentration. Consumer's accuracy is a measure of the spectral features that were correctly classified to correspond with the actual herbicide concentration that was applied. For example, herbicide concentration Z with a producer's accuracy of Y% and a consumer's accuracy of X% simply means that the system identified Y% of the spectral features as herbicide concentration Z, but only X% of the spectral features actually belonged with herbicide concentration Z.

Tables 3.1 to 3.4 each display a classification matrix for the hyperspectral data acquired 1, 4, 7, and 14 days after treatment (DAT) when glufosinate-ammonium was applied at various concentrations to susceptible corn. The classification matrix resulting from the hyperspectral data acquired 1 DAT (Table 3.1) through the ground-based spectral acquisition system displays an overall accuracy of 50%. Producer's accuracy results were greatest for the untreated and 1X rate of glufosinate-ammonium with accuracies of 94 and 97%, respectively. The system had difficulty classifying the spectral features associated with the two lowest rates of 1/32X and 1/16X, only providing producer's accuracies of 11 and 10%, respectively. The producer's accuracies for the remaining herbicide concentrations ranged from 34 to 52%. The consumer's accuracies ranged from 30 to 79% with the greatest consumer's accuracy being observed for the untreated.

Table 3.2 displays the classification matrix generated from the hyperspectral data recorded 4 DAT of glufosinate-ammonium. The system generated an overall accuracy 72% 4 DAT. The producer's accuracy of the untreated was 98% while 81% was observed for the lowest concentration of 1/32X. The lowest producer's accuracy of 44% was observed for the 1/16X concentration. The remaining intermediate (1/8X and 1/4X) concentrations received producer's accuracies of 61 and 79%, respectively. In addition, producer's accuracies of 72 and 63% were observed for the 1/2X and 1X rates. The consumer's accuracy for the 1/16X concentration was 94% while the remaining concentrations received consumer's accuracies of 59 to 83%.

The classification matrix resulting from the data acquired 7 DAT of glufosinate-ammonium is listed in Table 3.3. In this case, the overall accuracy generated by the system was 64%. Producer's accuracy results were greatest for untreated and the 1X concentration of glufosinate-ammonium with accuracies of 94 and 84%, respectively. Similar to what was observed 1 DAT, the system had difficulty correctly classifying the spectral features associated with the two lowest rates of 1/32X and 1/16X, only providing producer's accuracies of 34 and 38%. The producer's accuracies for the remaining herbicide concentrations ranged from 51 to 72%. The consumer's accuracies ranged from 47 to 88% with the greatest consumer's accuracy being observed for the 1/4X concentration.

Table 3.4 displays an overall accuracy of 59% of the classification matrix generated from the data acquired 14 DAT of glufosinate-ammonium. Producer's accuracies of 81, 76, and 79% with 52, 44, and 63 correct classifications of spectral features were observed for the untreated, 1/32X, and 1/4X concentrations, respectively. For the 1/2X and 1X concentrations, 63% producer's accuracy was observed. Only 11% producer's accuracy with 6 correctly classified spectral features was observed for the 1/16X concentration. Consumer's accuracies ranged from 47 to 77% for all concentrations.

Tables 3.5 to 3.8 display classification matrixes for the classification accuracies that were obtained 1, 4, 7, and 14 days after treatment (DAT) when clethodim was applied at various concentrations to corn. The classification matrix resulting from the hyperspectral data acquired 1 DAT of clethodim (Table 3.5) through the ground-based spectral acquisition system displays an overall accuracy of 43%. Producer's accuracy results were greatest for untreated, 1/4X, and 1/2X rates of clethodim with 62, 72, and 78 correct classifications, respectively. The system had difficulty correctly classifying the spectral features associated with the 1/16X rate, only providing producer's accuracies of 1% with only 1 correctly classified spectral feature. The producer's accuracies for the remaining two herbicide concentrations were 36 and 34% for the 1/32X and 1/8X rates, respectively. The consumer's accuracies ranged from 33 to 100% with the greatest consumer's accuracy being observed for the 1/16X rate, however, only one spectral feature was classified for this herbicide concentration.

Table 3.6 displays the classification matrix generated from the hyperspectral data recorded 4 DAT of clethodim. The system generated an overall accuracy of 51% 4 DAT. Producer's accuracy of the untreated was 93% with 77 correctly classified spectral features being associated with this treatment.

A producer's accuracy of 63% was observed for the 1/32X concentration with 41 spectral features being classified correctly. The lowest producer's accuracy of 18% was observed for the 1/16X concentration. The remaining concentrations received producer's accuracies of 37, 31, and 21%, respectively. Consumer's accuracy for the 1/16X concentration was 100% while the remaining concentrations received consumer's accuracies of 42 to 65%.

The classification matrix resulting from the data acquired 7 DAT of clethodim is listed in Table 3.7. In this case, the overall accuracy generated by the system was 62% with 45, 65, 24, 27, 37, and 66 correct classifications of the spectral features associated with the untreated, 1/32X, 1/16X, 1/8X, 1/4X, and 1/2X concentrations, respectively. Producer's accuracy results were greatest for the 1/32X and 1/2X concentrations of clethodim with 87 and 85% producer's accuracies. The producer's accuracies for the remaining herbicide concentrations ranged from 36 to 67%. The consumer's accuracies ranged from 46 to 92% with the greatest consumer's accuracy being observed for the untreated.

Table 3.8 displays an overall accuracy of 74% of the classification matrix generated from the data acquired 14 DAT of various clethodim concentrations. Producer's accuracies of 82, 80, 80, and 83% with 82, 68, 78, and 72 correct classifications of spectral features were observed for the untreated, 1/16X, 1/4X, and 1/2X concentrations, respectively. For the low concentration of 1/32X and intermediate concentration of 1/8X, producer's accuracies of 41 and 58% were observed, respectively. Consumer's accuracies ranged from 63 to 94% for all concentrations.

Tables 3.9 to 3.11 display classification matrixes for the classification accuracies that were obtained 4, 7, and 14 DAT when glyphosate was applied at 1/32X, 1/8X, and 1/2X concentrations to wheat. Data were not collectable 1 DAT for this experiment due to unfavorable weather conditions. The classification matrix resulting from the hyperspectral data acquired 4 DAT through the ground-based spectral acquisition system displays an overall accuracy of 73% (Table 3.9). Producer's accuracy results were greatest for the untreated with 85 correct classifications. The producer's accuracies decreased as glyphosate concentration decreased from 1/2X to 1/32X with accuracies of 79 to 56%, respectively. The consumer's accuracies ranged from 68 to 85% with the greatest consumer's accuracy being observed for the 1/32X concentration.

Table 3.10 displays the classification matrix generated from the hyperspectral data recorded 7 DAT of various glyphosate concentrations to wheat. The system generated an overall accuracy 86% 7 DAT. Producer's accuracy of the 1/2X concentration was 100% while the untreated, 1/8X, and 1/32X concentrations were 85, 73, and 86%, respectively. The observed consumer's accuracies were 90, 90, 71, and 97% for the untreated, 1/32X, 1/8X, and 1/2X concentrations.

The classification matrix resulting from the data acquired 14 DAT of clethodim is listed in Table 3.11. In this case, the overall accuracy generated by the system was 92% with 87, 89, 77, and 78 correct classifications of the spectral features associated with the untreated, 1/32X, 1/8X, and 1/2X concentrations, respectively. A producer's accuracy of 98% was observed for untreated. The producer's accuracies for the remaining herbicide concentrations (1/32X, 1/8X, and 1/2X) were 84, 94, and 94%. The consumer's accuracies ranged from 83 to 100% with the greatest accuracy being observed for the 1/2X concentration.

Visual injury ratings recorded 7, 14, and 28 DAT resulted in an expected stair-step pattern with the visual injury decreasing as herbicide concentration decreased, regardless of specific herbicide or the crop it was applied to (Table 3.12). Visual injury was significantly greater than the untreated for all rates of glufosinate-ammonium 7 DAT, with the exception of the 1/32X rate. However, 14 and 28 DAT, all glufosinate-ammonium rates provided significant injury when compared to the untreated, although the level of injury did decrease over time. Similarly, visual injury ratings recorded 7 DAT of various rates of clethodim were applied to corn showed a significant increase in injury when compared to the untreated, with the exception of the two lowest concentrations. Again, 14 and 28 DAT, significant injury was observed for all rates of clethodim when compared to the untreated. Significant injury was observed 7 and 14 DAT when glyphosate concentrations of 1/8X and 1/2X were applied to wheat. However, by 28 DAT, no visual injury was observed for any rate. The 1/32X rate provided no visual injury 7, 14, or 28 DAT of glyphosate to wheat.

Crop yield reductions expressed as a percentage of the yield potential of the untreated are displayed in Table 3.13. No reduction in crop yield was observed after glufosinate-ammonium was applied at 1/32X, 1/16X, 1/8X, and 1/4X concentrations to susceptible corn. A 30% yield reduction was observed when a 1/2X rate of glufosinate-ammonium was applied, however, this was not significant when compared to the untreated. A significant yield reduction of 72% was observed for the 1X rate of glufosinate ammonium. After clethodim concentrations were applied to corn, a 19, 27, and 30% reduction in crop yield was observed for the 1/32X, 1/16X, and 1/8X rates, respectively. However, these reductions in crop yield were not found to be significant when compared to the untreated. Significant reductions in corn yield of 60 and 100% were observed for the 1/4X and 1/2X rates of clethodim. A reduction of 5% in wheat yield was observed following application of glyphosate at the 1/32X concentration, however, this was not significant when compared to the untreated. Significant reductions of 12 and 31% in wheat yield was observed for the 1/8X and 1/2X rates of glyphosate, respectively.

Conclusion

These data indicate that the predictive capability of this system varies for correctly classifying herbicide concentrations through spectral features. The generated overall accuracies of the systems' capability for classification was highest 4 DAT when glufosinate-ammonium was applied to susceptible corn and 14 DAT when clethodim was applied to corn and glyphosate to wheat. This can be expected as the spectral response of the plants will likely vary after applications of these specific herbicides. For example, plants exposed to a labeled rate of glufosinate-ammonium will exhibit necrosis of leaves and young shoots within 2 to 4 days after the application if conditions are favorable (Anonymous 2012a). Plants exposed to clethodim, however, will generally show symptoms 7 to 14 days after an application of the labeled rate (Anonymous

2012b). When susceptible plants are exposed to glyphosate, symptoms are sometimes visible within 2 to 4 days for some species, but can take 7 or more days for symptoms to occur (Anonymous 2012c). In addition, the specimen label for glufosinate ammonium indicates that an additional application at the labeled rate may be required to effectively control corn (Anonymous 2012a). This indicates that corn exposed to sub-lethal concentrations of glufosinate-ammonium may recover and continue normal growth. Therefore, these overall classification accuracies mimic what would be expected from applications of these herbicides to susceptible plant species.

Specifically, the predictive capability of this system for classifying spectral features associated with concentrations of glufosinate-ammonium would be expected to be higher immediately after application when compared to two weeks after the application. In addition, producer's accuracies 1 DAT would likely be greater for higher concentrations or the untreated, as was seen in these data, due to the fact that the herbicide will not likely have taken affect at sub-lethal concentrations. At 4 DAT, these data indicate that lower producer's accuracies were observed for the intermediate and higher rates. This may be expected as the plants are exhibiting similar symptoms, regardless of rate, which can be confirmed with the similar levels of visual injury that were noted (Table 3.12). The overall accuracies decreased from 64% 7 DAT to 59% 14 DAT. This could be due to the corn recovering from lower concentrations and beginning to grow normally.

These data indicate that it is likely that the predictive capability of this system for classifying spectral features associated with concentrations of clethodim applied to corn and glyphosate to wheat will increase over time. In other words, with herbicides that require a longer period of time to affect the plant, it may be more difficult to discern a noticeable difference in spectral features associated with specific herbicide concentrations immediately after exposure. In both of these cases, the highest overall accuracy was observed 14 DAT.

As described through these data, it is possible to utilize spectral features to classify certain concentrations of herbicides which may influence a crop after an off-target herbicide deposition instance. These data indicate that this system is capable of predicting spectral features associated with multiple herbicides. In doing so, it is possible that in classifying spectral features associated with multiple herbicides that either work rapidly within the plant or require a week or longer to develop symptoms, this system could be utilized to not only express the level of injury which may occur with off-target herbicide deposition, but also provide information regarding yield losses which may or may not occur. This would be a great benefit for the producers whose crops are affected by off-target herbicide deposition. This could allow for proper decisions to be made regarding the overall health of the current crop and whether or not the level of injury presents a financial liability should the crop be left as is.

Table 3.1 Classification matrix resulting from data generated through a multi-classifier decision fusion of hyperspectral data obtained through a ground-based spectral acquisition system 1 day after treatment of various rates of glufosinate-ammonium applied to susceptible corn.

		Herbicide Concentration[1]							
		Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	1X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	99	0	0	0	0	1	5	94%
	1/32X	11	9	4	27	3	3	25	11%
	1/16X	3	1	8	18	9	4	36	10%
	1/8X	8	2	0	50	9	1	27	52%
	1/4X	2	0	0	6	30	17	34	34%
	1/2X	3	0	0	1	3	37	39	45%
	1X	0	0	0	0	0	2	72	97%
	Consumer's Accuracy[3]	79%	75%	67%	49%	56%	57%	30%	50%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.59 kg ai/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

Table 3.2 Classification matrix resulting from data generated through a multi-classifier decision fusion of hyperspectral data obtained through a ground-based spectral acquisition system 4 days after treatment of various rates of glufosinate-ammonium applied to susceptible corn.

		Herbicide Concentration[1]							
		Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	1X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	85	1	0	0	0	1	0	98%
	1/32X	8	66	1	3	1	2	0	81%
	1/16X	26	6	33	5	1	4	0	44%
	1/8X	4	10	1	50	14	3	0	61%
	1/4X	3	0	0	3	70	9	4	79%
	1/2X	6	0	0	1	11	66	8	72%
	1X	3	0	0	0	5	27	59	63%
	Consumer's Accuracy[3]	63%	80%	94%	81%	69%	59%	83%	72%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.59 kg ai/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

Table 3.3 Classification matrix resulting from data generated through a multi-classifier decision fusion of hyperspectral data obtained through a ground-based spectral acquisition system 7 days after treatment of various rates of glufosinate-ammonium applied to susceptible corn.

		Herbicide Concentration[1]							
		Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	1X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	60	0	0	2	0	2	0	94%
	1/32X	8	18	5	2	1	15	4	34%
	1/16X	6	2	22	17	0	10	1	38%
	1/8X	2	3	3	52	2	14	4	65%
	1/4X	1	2	1	9	28	9	5	51%
	1/2X	3	1	0	1	1	53	15	72%
	1X	0	0	0	1	0	10	58	84%
	Consumer's Accuracy[3]	75%	69%	71%	62%	88%	47%	67%	64%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.59 kg ai/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

Table 3.4 Classification matrix resulting from data generated through a multi-classifier decision fusion of hyperspectral data obtained through a ground-based spectral acquisition system 14 days after treatment of various rates of glufosinate-ammonium applied to susceptible corn.

Herbicide Concentration[1]	Herbicide Concentration[1] →	Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	1X	Producer's Accuracy[2]
	Untreated	52	4	4	2	2	0	0	81%
	1/32X	8	44	1	3	2	0	0	76%
	1/16X	5	7	6	16	22	1	0	11%
	1/8X	8	16	0	24	19	1	0	35%
	1/4X	4	5	0	6	63	2	0	79%
	1/2X	0	0	0	0	15	45	12	63%
	1X	0	1	0	0	0	23	41	63%
	Consumer's Accuracy[3]	68%	57%	55%	47%	51%	63%	77%	59%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.59 kg ai/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

Table 3.5 Classification matrix resulting from data generated through a multi-classifier decision fusion of hyperspectral data obtained through a ground-based spectral acquisition system 1 day after treatment of various rates of clethodim applied to susceptible corn.

		Herbicide Concentration[1]						
		Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	62	8	0	0	11	9	69%
	1/32X	0	34	0	8	41	11	36%
	1/16X	0	3	1	7	13	50	1%
	1/8X	0	23	0	32	33	5	34%
	1/4X	4	9	0	13	72	5	70%
	1/2X	1	2	0	6	18	78	71%
	Consumer's Accuracy[3]	75%	43%	100%	41%	33%	36%	43%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.10 kg ai/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

Table 3.6 Classification matrix resulting from data generated through a multi-classifier decision fusion of hyperspectral data obtained through a ground-based spectral acquisition system 4 days after treatment of various rates of clethodim applied to susceptible corn.

		Herbicide Concentration[1]						
		Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	77	2	0	1	0	0	93%
	1/32X	17	41	0	5	1	0	63%
	1/16X	16	5	13	2	20	3	18%
	1/8X	14	6	0	27	4	0	37%
	1/4X	27	12	0	6	28	1	31%
	1/2X	11	5	0	2	8	20	21%
	Consumer's Accuracy[3]	45%	57%	100%	60%	42%	65%	51%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.10 kg ai/ha.

[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.

[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.

[4] – Overall accuracy of the system.

Table 3.7 Classification matrix resulting from data generated through a multi-classifier decision fusion of hyperspectral data obtained through a ground-based spectral acquisition system 7 days after treatment of various rates of clethodim applied to susceptible corn.

		Herbicide Concentration[1]						
		Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	45	8	0	7	4	3	67%
	1/32X	0	65	0	1	6	3	87%
	1/16X	1	0	24	8	3	6	53%
	1/8X	2	16	0	27	19	10	36%
	1/4X	1	10	0	10	37	12	49%
	1/2X	0	0	1	2	3	66	85%
	Consumer's Accuracy[3]	92%	66%	75%	49%	51%	46%	62%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.10 kg ai/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

Table 3.8 Classification matrix resulting from data generated through a multi-classifier decision fusion of hyperspectral data obtained through a ground-based spectral acquisition system 14 days after treatment of various rates of clethodim applied to susceptible corn.

		Herbicide Concentration[1]						
		Untreated	1/32X	1/16X	1/8X	1/4X	1/2X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	82	0	8	3	5	2	82%
	1/32X	27	31	9	2	5	1	41%
	1/16X	12	0	68	1	3	1	80%
	1/8X	5	2	4	49	23	2	58%
	1/4X	4	0	1	1	78	12	80%
	1/2X	0	0	1	0	8	72	83%
	Consumer's Accuracy[3]	63%	94%	75%	88%	64%	70%	74%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.10 kg ai/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

Table 3.9 Classification matrix resulting from data generated through a multi-classifier decision fusion of hyperspectral data obtained through a ground-based spectral acquisition system 4 days after treatment of various rates of glyphosate applied to wheat.

		Herbicide Concentration[1]				
		Untreated	1/32X	1/8X	1/2X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	107	1	6	1	93%
	1/32X	2	60	18	28	56%
	1/8X	33	4	70	4	63%
	1/2X	8	6	9	85	79%
	Consumer's Accuracy[3]	71%	85%	68%	72%	73%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.86 kg ae/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

Table 3.10 Classification matrix resulting from data generated through a multi-classifier decision fusion of hyperspectral data obtained through a ground-based spectral acquisition system 7 days after treatment of various rates of glyphosate applied to wheat.

		Herbicide Concentration[1]				
		Untreated	1/32X	1/8X	1/2X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	92	2	14	0	85%
	1/32X	3	77	23	2	73%
	1/8X	7	7	90	1	86%
	1/2X	0	0	0	98	100%
	Consumer's Accuracy[3]	90%	90%	71%	97%	86%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.86 kg ae/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

Table 3.11 Classification matrix resulting from data generated through a multi-classifier decision fusion of hyperspectral data obtained through a ground-based spectral acquisition system 14 days after treatment of various rates of glyphosate applied to wheat.

		Herbicide Concentration[1]				
		Untreated	1/32X	1/8X	1/2X	Producer's Accuracy[2]
Herbicide Concentration[1]	Untreated	87	0	2	0	98%
	1/32X	3	89	14	0	84%
	1/8X	2	3	77	0	94%
	1/2X	2	3	0	78	94%
	Consumer's Accuracy[3]	93%	94%	83%	100%	92%[4]

[1] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.86 kg ae/ha.
[2] – Percent accuracy of the system's capability for correctly classifying the spectral feature(s) that correspond with the specific herbicide concentration.
[3] – Percent of the spectral feature(s) which were correctly classified to correspond with the actual herbicide concentration that was applied.
[4] – Overall accuracy of the system.

Table 3.12 Percent visual injury ratings collected after application of various rates of herbicides to susceptible corn.

	Herbicide Drift to Susceptible Corn						Herbicide Drift to Wheat		
	Glufosinate-ammonium			Clethodim			Glyphosate		
	Days After Treatment[1]								
	7	14	28	7	14	28	7	14	28
Application Rate[2]	---------------%---------------			---------------%---------------			---------------%---------------		
Untreated	0	0	0	0	0	0	0	0	0
1/32X	7	10	11	0	0	0	0	0	0
1/16X	18	16	14	2	13	23	--[3]	--[3]	--[3]
1/8X	23	20	20	5	28	22	7	16	0
1/4X	35	33	27	12	50	38	--[3]	--[3]	--[3]
1/2X	62	62	49	19	63	82	23	35	0
1X	74	75	64	--[3]	--[3]	--[3]	--[3]	--[3]	--[3]
LSD (0.05)[4]	9	9	14	4	6	16	4	4	NS

[1] – Visual ratings recorded 7, 14, and 28 days after various rates of the listed herbicides were applied to corn.
[2] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.59 kg ai/ha, clethodim rate of 0.10 kg ai/ha, and glyphosate rate of 0.86 kg ae/ha.
[3] – Herbicide concentrations not included in this experiment.
[4] – Means separated according to Fisher's Protected LSD at $P = 0.05$.

Table 3.13 Percent crop yield reductions after simulated herbicide drift applications.

	Herbicide Drift to Susceptible Corn		Herbicide Drift to Wheat
	Glufosinate-ammonium	Clethodim	Glyphosate
Application Rate[2]	Yield Reduction[1]		
	-----%-----		
Untreated	0	0	0
1/32X	0	19	5
1/16X	0	27	--[3]
1/8X	0	30	12*
1/4X	0	60*	--[3]
1/2X	30	100*	31*
1X	72*	--[3]	--[3]

[1] – Crop yield reductions expressed as a percentage of the untreated after various rates of the listed herbicides were applied to corn.
[2] – Herbicide concentration expressed as a fraction of the labeled glufosinate-ammonium application rate of 0.59 kg ai/ha, clethodim rate of 0.10 kg ai/ha, and glyphosate rate of 0.86 kg ae/ha.
[3] – Herbicide concentrations not included in this experiment.
* – Means separated according to Fisher's Protected LSD at $P = 0.05$.

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CHAPTER IV

DIFFERENTIATION BETWEEN CROP STRESS INDUCED FROM HERBICIDE APPLICATION AND STRESS DUE TO NUTRIENT OR MOISTURE DEFECIENCY

Introduction

Crop stress is a major concern for producers. Stress can influence a crop’s growth and development and have negative impacts on yield. There are many types of stresses including but not limited to herbicide stress, nutrient stress, and drought stress. Herbicide induced stress on a crop is not new. Typically, stress caused by herbicides occurs during an off-target deposition instance of a herbicide application. The Environmental Protection Agency (EPA) defines spray drift as “The physical movement of a pesticide through air at the time of application or soon thereafter, to any site other than that intended for application (often referred to as off-target)” (Environmental Protection Agency 2009). There are many variables which can influence off-target deposition. These factors include environmental conditions at time of application (i.e. wind speed, temperature, and humidity), herbicide formulation, application pressure, application speed, boom height, nozzle type, and droplet size (Carlsen et al. 2006).

With increased acreage being planting in varieties/hybrids that contain herbicide resistant traits, herbicide injury to non-target crops continues to cause problems for producers. As the industry continues to develop new genetic traits that will allow multiple herbicide chemistries to be applied safely to the target crop, it is likely that herbicide induced stress will continue to be a major concern in non-target fields. To date, commercially available herbicide-tolerant varieties/hybrids contain traits which allow herbicides like glyphosate and glufosinate-ammonium to be applied as a broadcast application during the growing season. Both glyphosate and glufosinate-ammonium are broad spectrum herbicides which have activity on a large number of plant species, including major crops like non-transgenic soybeans, corn, cotton, and wheat. Glyphosate was applied to 68 and 66% of U.S. cotton and corn acres in 2010, respectively (USDA NASS 2012).These applications totaled approximately 68 million pounds of glyphosate applied in the U.S. in 2010. Therefore, it is possible for crop stress from herbicide applications to occur in neighboring fields that contain susceptible crop varieties/hybrids.

Previous research focusing on herbicide stress has shown that ultra-low rates of glyphosate can reduce corn yield (Rowland 2000). Experiments conducted to simulate glyphosate drift in corn showed yield reductions of 78, 43, and 22% for simulated applications of 140, 70, and 35 grams acid equivalent per hectare (g ae/ha) of glyphosate, respectively (Ellis et al. 2003). Roider et al. (2007) found a 43% decrease in wheat yield when glyphosate was applied at 70 grams active ingredient per hectare (g ai/ha), which is approximately 6% of the normal use rate for this herbicide. Ellis et al. (2003) found that height reductions and foliage discoloration from sub-lethal applications of glyphosate to both rice and corn crops were minimal, but negative effects on yields were significant. Stress caused by lack of available moisture or nutrients can also cause reductions in crop yield. Previous research has shown significant reductions in soybean yield when water stress at any level was imposed during the reproductive phase of the crop (Kirnak et al. 2008). Clay et al. (2006) observed reductions in corn yield when both nitrogen and moisture stress were imposed to corn.

Technological advances have allowed new or improved methods for producer's to monitor the condition of their crops during the growing season. Producers are able to utilize technologies such as internet, mobile phones, global positioning systems, site-specific applicators, and remotely sensed data to assist with decision making pertaining to best management practices which can improve crop production and provide protection to the environment. Producers who are adopting this technology can combine many of these tools to assess equipment performance, monitor environmental conditions, and evaluate the condition of their crops at any given time. Remote sensing tools are of particular interest with the practice of precision agriculture.

Tools which collect remotely sensed data, such as multispectral and hyperspectral sensors, can be utilized as a means to assess specific conditions within a given field which relate to crop yield (Seidl et al. 2004). For example, crop stress induced by pests, moisture or nutrient availability, or the crops reaction to a specific management practice can be monitored with remote sensing tools (Tartachnyk et al. 2006, Lichtenthaler 1996). Therefore, remote sensing systems have proven to be useful for many applications in production agriculture. These applications include detecting crop stress from lack of nutrients and moisture in crops such as corn and wheat (Barnes et al. 2000, Ghulam et al. 2008, Barker and Sawyer 2010), weed infestations (Chang et al. 2004), yield performance (Chang et al. 2003), crop stand density (Thorp et al. 2008), diseases in wheat (Franke and Menz 2007), and injury from herbicide applications (Thelen et al. 2004, Henry et al. 2004). Ground-based systems, aerial imagery, and satellite imagery are options for obtaining remotely sensed data. Everman et al. (2008) utilized both a handheld spectroradiometer and aerial imagery to evaluate the effects of herbicides on the spectral reflectance of corn. Satellite imagery has been used to measure the nutrient uptake of winter cover crops (Hively et al. 2009). The use of such systems can provide valuable information about plant health from reflectance measurements of the crop canopy.

Nitrogen and moisture stress as well as stress caused by herbicide applications can interact to influence yields in agricultural fields. Remotely sensed data can be used as a tool to assess these stresses (Barnes et al. 2000). The theory behind the utilization of remote sensing to detect plant stress is based on the assumption that stress is interfering with photosynthetic reactions within the plant or the physical structure of the plant. Therefore, stress can affect the absorption of energy from light which changes the reflectance of energy from the plants (Riley et al. 1989, Hatfield and Pinter 1993). The nitrogen content found in leaves of many crops is an important indicator of growth status, quality, and yield (Cui et al. 2009). Nitrogen stress reduces the amount of chlorophyll and can result in increased reflectance of photosynthetically active light (Clay et al. 2006) and decreased reflectance in the near-infrared light (Cui et al. 2009, Yoder and Pettigrew-Crosby 1995). Water stress can influence reflectance due to reduced photochemical activity of chlorophyll (Clay et al. 2006, Souza et al. 2004). Herbicide applications to sensitive crops have also shown variations in reflectance. A reduction in NIR reflectance in corn was found when glyphosate was applied at 0.433 kg ae/ha, which is approximately 50% of the normal use rate for this herbicide (Irby 2009). Vegetation has unique characteristics regarding solar irradiance. Reflectance in the visible light spectrum (400-700 nm) is very low, transmittance is zero, and absorptance is high (Thelen et al. 2004). In the near-infrared (NIR) portion of the spectrum (700-1350 nm), both reflectance and transmittance are high and absorptance is low (Thelen et al. 2004). Because of these characteristics of reflectance of vegetation, multiple spectral vegetation indices have been developed. The normalized difference vegetation index (NDVI) is commonly used as an indication of plant vigor. NDVI is the ratio of NIR-Red/NIR+Red. Clay et al. (2006) used NDVI to measure water and nitrogen stress in corn while Henry et al. (2004) used NDVI to classify herbicide injury to soybeans and corn. Crop water stress indices (CWSI) have been used to map water stress in crops. This index uses canopy temperature and environmental conditions to calculate a value which describes water stress on a scale from 0 to

1, with 0 being no water stress and 1 being complete water stress. Barnes et al. (2000) used the CWSI paired with NDVI to map water stress in cotton. Experiments have shown that hyperspectral vegetation indices can provide a more accurate assessment of crop parameters than did equivalent indices from multispectral sensors (Thenkabail et al. 2002). However, hyperspectral data are more complicated when compared to multispectral data due to the volume of data which is obtained (Karimi et al. 2005). In order to extract useful information from larger hyperspectral data sets, it is important to first select a range of bands that can be used to describe information about the specific target.

Remotely sensed data can provide valuable information about the overall health of a plant in instances where crop injury is suspected but not visible. Some factors that should be considered when using remotely sensed data in this capacity include time of day/year, topography, soil type, and crop type. Previous research has shown that reflectance values in tilled fields were primarily influenced by soil characteristics during the early stages of the growing season (Huete et al. 1985). Chang et al. (2003) reported that characteristics of spectral reflectance are influenced by plant factors more than soil factors as the growing season progresses. This is expected due to the methods by which many of our agricultural crops are grown. Typically, row crops are planted in a variety of spacings both between the rows as well as within each row. These spacings are chosen based on the optimum plant population which will maximize yield and provide canopy cover to efficiently use light energy in the plant’s photosynthetic processes. Therefore, as the season progress, the crop canopy begins to cover the soil surface allowing spectral reflectance to be influenced more by plant factors rather than soil factors.

The ability to rapidly detect and assess herbicide induced stress to a crop would be beneficial in many aspects. From a producer's standpoint, a rapid response time is needed in order to make a management decision about the stressed crop. In the event of herbicide injury to a crop, producers could use the information obtained from the remotely sensed data coupled with data showing yield reductions correlated to reflectance measurements to make informed decisions for replanting or leaving the injured crop in the field. This type of situation is often the most difficult decision that a producer might have to make. Factors such as the type of injury and time of year play important roles in this process. Having tools in place which can assist in rapidly making these decisions could allow producers to salvage their investment rather than accepting a total loss. A commonly asked question while using remote sensing tools in the event of off-target deposition; however, is whether or not it is actually herbicide stress affecting the observed changes in reflectance. In other words, is it truly the herbicide stress causing changes in reflectance or is it another factor such as moisture or nutrient stress. Therefore, this research was conducted in order to compare reflectance values obtained after herbicide, nutrient, and moisture stress was induced to corn to discern if differences exist in specific wavelengths that best correlate with the respective stresses.

Materials and Methods

This experiment was conducted in a greenhouse environment at the R.R. Foil Plant Science Research Center near Starkville, MS. The experiment was replicated twice. Plants were grown in 3,800 cubic centimeter (cm^3) pots measuring 15 centimeters (cm) in diameter and 19 cm in height. The pots were filled with masonry sand containing the appropriate levels of macro- and micro-nutrients for corn production, with the exception of the target deficiency nutrient of nitrogen. Pioneer P1184HR corn seed was planted with 2 seeds per pot and thinned to 1 plant per pot after corn emergence. The air temperature in the greenhouse was maintained at 29° C during a 14-hour day and 18° C during the dark period. Treatments included three herbicide rates, three nitrogen rates, three moisture rates, and an untreated check for comparison purposes. Each treatment was replicated 4 times. The herbicide treatments were based off the labeled rate of 0.10 kg ai/ha of clethodim, the active ingredient in the herbicide Select Max®. The actual rates of clethodim included concentrations of 0.05, 0.013, and 0.0016 kg ai/ha, which correspond to 1/2X, 1/8X, and 1/64X fractions of the recommended labeled rate. Herbicide treatments were applied at the V3 growth stage. Treatments for the various moisture and nitrogen rates were applied at the beginning of the experiment. In order to simulate a nutrient deficiency, three rates of nitrogen were mixed with the masonry sand while all remaining macro- and micro-nutrients were held constant. The three levels of nitrogen consisted of a normal N rate of 90 kg/ha with medium and low rates of 60 and 30 kg N/ha, respectively. This normal rate was selected as an appropriate level of starter N to be applied for a dry-land corn yield goal of 11,500 kg/ha. Similarly, the three moisture treatments received varying amounts of deionized water twice daily to maintain soil moisture content at low (wilting point), medium, and normal (field capacity) levels. Ultimately, the herbicide treatments were maintained at normal nutrient and water levels, the nitrogen treatments received no herbicide and normal water levels, and the moisture treatments received no herbicide and normal nutrient levels. In addition, the untreated check received no herbicide treatment, normal moisture, and normal nutrients in order to maintain this treatment as a healthy control for comparison purposes. Data collection consisted of soil moisture readings in the form of percent volumetric water content collected using a portable soil moisture sensor, leaf chlorophyll content readings of a unitless value between 0 and 50 using a SPAD chlorophyll meter, plant heights in centimeters (cm), and leaf clip reflectance measurements using the Analytical Spectral Devices (ASD™) Fieldspec Pro handheld spectroradiometer. Volumetric water content, leaf chlorophyll content, and plant height data were recorded prior to herbicide application as well as 1, 3, 7, and 14 days after herbicide treatment (DAT) which correspond to 16, 18, 22, and 29 days after corn emergence (DAE). Spectral data were acquired 3, 7, and 14 DAT of the herbicide induced stress. A Normalized Difference Vegetation Index (NDVI) was calculated from the spectral data 3, 7, and 14 DAT. Volumetric water content, leaf chlorophyll content, plant height data, and NDVI were averaged across both experiments and subjected to an analysis of variance with means separated using Fisher’s Protected LSD at P = 0.05. Spectral data were combined over both experiments to evaluate specific wavelengths which correlate to herbicide, nutrient, or moisture stresses. Correlation coefficients were generated to determine which spectral features were correlated with the various forms of stress.

Results and Discussion

Data were first analyzed in order to determine if the experiment was successful for separating stresses induced by the various treatments of moisture and nutrient deficits and herbicide rates. Both moisture stress and nutrient stress simulated by deficit of soil nitrogen were subjected to the experiment at planting. Data collection did not begin until herbicide stress in the form of sub-lethal concentrations of clethodim was induced to the corn at the V3 growth stage. A period of 15 days passed from corn emergence to induction of herbicide stress. By the end of the sampling time, corn had reached the V4 growth stage. Therefore, data for moisture and nitrogen stress are displayed in terms of days after corn emergence (DAE) while data for herbicide stress are displayed as days after herbicide treatment (DAT).

Table 4.1 lists the results of the data for plant height, leaf chlorophyll content, and volumetric water content. Prior to herbicide application, no differences in plant height were observed. A significant difference in plant height was observed for the low level of moisture when compared to the untreated with height reductions of 6.2, 6.5, 6.4, and 6.3 cm 16, 18, 22, and 29 DAE, respectively. Plant height reductions were present for both the 1/2X and 1/8X clethodim rates at 3, 7, and 14 DAT. A reduction in plant height of 5 cm was also observed for the medium moisture level 18 DAE.

Significant reductions in leaf chlorophyll content were observed for the medium and low levels of nitrogen prior to herbicide application (Table 4.1) with reductions of 3.7 and 5.4, respectively. The same levels of nitrogen showed numerical differences 16 DAE, however, only the low nitrogen level was found to be significant. By 18 DAE (3 DAT), reductions in leaf chlorophyll content of 4.5 and 6.6 were observed for the 1/2X concentration of clethodim and low level of moisture stress, respectively. In addition, leaf chlorophyll content reductions were observed for both the medium and low levels of nitrogen 18 DAE. Significant reductions in leaf chlorophyll content were also observed for all herbicide concentrations as well as the medium and low levels of nitrogen and moisture at 7 and 14 DAT (22 and 29 DAE).

With respect to volumetric water content, moisture treatments were applied twice daily in order to maintain soil moisture content at low, medium, and sufficient levels. Significant reductions in volumetric water content were only observed where moisture deficit was designed to be at the medium and low levels. Reductions ranging from 5.4 to 6.1% were observed for the medium moisture level and 10.4 to 10.7% for the low moisture level across the 5 sampling times (Table 4.1).

NDVI values were calculated for the spectral data collected 3, 7, and 14 DAT (18, 22, and 29 DAE) (data not shown). NDVI values were calculated using a range of 800 to 900 nm for the NIR and 600 to 700 for the Red. No significant differences in NDVI were observed for any treatment.

The results from the analysis of plant height, leaf chlorophyll content, and volumetric water content indicate that the experiment was successful for subjecting corn to various levels of stress between herbicide, nitrogen deficiency, and moisture deficit. As would be expected with herbicide stress, plant height reductions began to occur 3 DAT and continued for the remaining period of data sampling. In addition, when corn is subjected to moisture deficit during the early vegetative stages, plant height reductions can occur (Olaoye et al. 2009). This phenomenon was observed in this experiment as reductions in plant height were present for the low level of moisture 16, 18, 22, and 29 DAE. With respect to nitrogen deficiency, reductions in leaf chlorophyll content were observed both prior to as well as during the 2 week sampling time for the low level of nitrogen. Reductions in leaf chlorophyll content were also observed for the medium level of nitrogen during this same period, with the exception of 16 DAE. Also, reductions in leaf chlorophyll content occurred 7 and 14 DAT (22 and 29 DAE) for all levels of induced herbicide and moisture stress. Reductions in leaf chlorophyll content were also observed for the highest levels of induced herbicide and moisture stress 3 DAT (18 DAE). These observations coupled with the expected differences in volumetric water content indicate that different levels of stress within each of the three categories were influencing normal plant growth and development.

After confirming that the different types of stress were indeed present, the hyperspectral data were analyzed in order to determine which wavelength(s) may or may not be indicative of each type of stress in order to determine if a specific region of the electromagnetic spectrum can be used to separate these particular forms of plant stress. Correlations were made between spectral features associated with the most stressful treatments of herbicide and nutrient stress, herbicide and moisture stress, and nutrient and moisture stress 3, 7, and 14 DAT of herbicide stress induction (18, 22, and 29 DAE). The correlations between spectral features and stress type are displayed in Figures 4.1 to 4.9. Correlation coefficients were generated comparing the pairs of stress types to determine a linear relationship between each pair of stress types. This relationship was used to determine the correlation of spectral features (range within the electromagnetic spectrum) and stress type (positive vs. negative correlation).

In terms of wavelength response to herbicide and nutrient stress, the observed correlation coefficients indicate that herbicide stress has more impact on the visible and NIR portions of the spectrum both 3 and 7 DAT (18 and 22 DAE) (Figures 4.1 and 4.2) while nutrient stress is correlated to the infrared (IR). However, by 14 DAT (29 DAE), nutrient stress seems to correlate more to the visible and NIR portions of the spectrum while herbicide stress is correlated to the IR portion (Figure 4.3). This is likely due to the fact that at 14 DAT, the herbicide has caused severe injury to the plant as would be expected due to the fact that plants exposed to clethodim will generally show symptoms 7 to 14 days after an application of the labeled rate (Anonymous 2012).

Graphs displaying the correlation coefficients of spectral bands associated with herbicide and moisture stress are shown in Figures 4.4 to 4.6. Correlation coefficients of spectral bands indicate that the NIR region of the electromagnetic spectrum is correlated to herbicide stress 3 DAT (18 DAE) while the visible and IR portions of the spectrum are correlated to moisture stress (Figure 4.4). However, by 7 DAT (22 DAE), moisture stress was correlated to the entire range of wavelengths (Figure 4.5). Correlation coefficients of spectral bands calculated 14 DAT (29 DAE) show that herbicide stress was correlated to the blue (400 to 500 nm), green (500 to 600 nm), and NIR regions of the spectrum while moisture stress was correlated to the red (600 to 700 nm) and IR regions (Figure 4.6).

The observed correlation between wavelengths associated with nutrient and moisture stress 18 DAE express similar spectral response as was observed for herbicide and moisture stress at this same time. Correlation coefficients of wavelengths associated with these two forms of stress indicate that the NIR region of the electromagnetic spectrum is correlated to nutrient stress 18 DAE while the visible and IR portions of the spectrum are correlated to moisture stress (Figure 4.7). Both 22 and 29 DAE, however, the data indicate that moisture stress is best correlated with the visible and NIR regions of the spectrum while nutrient stress is best correlated with the IR regions (Figures 4.8 and 4.9).

Classification matrixes were also generated in order to determine classification accuracies for spectral features associated with the various forms of stress (data not shown). Overall accuracies were very low ranging from 13.8 to

16.3%. The low overall accuracies were likely due to the limited amount of hyperspectral data which were available.

Conclusion

The results of this experiment demonstrate a response in spectral reflectance from a corn crop under moisture and nitrogen stress at the time of an off-target herbicide deposition instance. These data indicate varying results in terms of correlating a range of wavelengths that may be capable of separating these specific forms of crops stress. At 3 days after the simulated off-target herbicide deposition instance (18 DAE), these data indicate that the visible to NIR range of the spectrum is best suited to identify herbicide stress as long as moisture stress is not present (Table 4.2). If moisture stress is present, then only the NIR portion of the spectrum is suited to characterize herbicide stress (Table 4.2). Conversely, if herbicide stress is not present, then the NIR region is best suited to identify nutrient stress when it is present with moisture stress (Table 4.2). Observations at 7 DAT (22 DAE) indicate that the visible to NIR region of the spectrum is best for identifying herbicide stress as long as moisture stress is not present (Table 4.3). However, if moisture stress is present simultaneously with herbicide and nitrogen stress, the visible to NIR range of the spectrum is more useful for identifying stress related to moisture deficit (Table 4.3). By 14 DAT (29 DAE) of the simulated off-target herbicide deposition instance, the spectral data indicate that the visible to NIR region of the spectrum can be used to better identify stress related to nitrogen deficiency than to herbicide stress Table 4.4). However, this same portion of the spectrum is still capable of identifying herbicide stress compared to moisture stress, as long as nutrient deficiency is not present (Table 4.4). If herbicide stress is not present, then moisture stress is better identified with the visible to NIR region of the spectrum compared to nitrogen stress (Table 4.4).

While these data indicate that remotely sensed data are capable of identifying stress related to crops, it seems unlikely that specific stress types can be identified with the equipment used in this experiment if multiple forms of stress are present. This experiment was conducted in a controlled greenhouse environment. In a true cropping scenario, many variables may be present which interact to influence spectral reflectance. Without knowing the specific form of crop stress, it would be difficult to utilize remotely sensed data to accurately quantify negative impacts on the crop. However, if moisture stress were removed as may be the case in an irrigated cropping system, these data demonstrate that the visible and NIR regions of the electromagnetic spectrum can be used to separate stress related to off-target herbicide deposition when present with stress in the form of nitrogen deficiency. This knowledge could prove useful in the event of off-target herbicide deposition to a crop which is deficient in nitrogen. This could allow for proper decisions to be made regarding the overall health of the current crop and whether or not the level of injury presents a financial liability should the crop be left as is.

Table 4.1 Plant height, leaf chlorophyll content, and volumetric water content recorded before as well as 1, 3, 7, and 14 days after herbicide stress was induced to corn.

		Plant Height					Leaf Chlorophyll Content					Volumetric Water Content				
		Days After Treatment														
		0	1	3	7	14	0	1	3	7	14	0	1	3	7	14
		--------------------cm--------------------										--------------------%--------------------				
H[1]	1/2X	24.3	27.1	26.3	26.8	27.9	29.2	30.4	26.5	21.5	11.1	15.0	15.0	14.8	15.0	15.0
	1/8X	23.9	27.0	26.4	28.9	30.3	31.4	35.0	28.3	28.3	21.6	15.1	15.0	14.9	15.0	14.8
	1/64X	25.9	27.8	31.6	35.0	43.6	30.5	33.3	27.6	29.4	15.6	15.0	14.9	15.0	14.8	14.7
M[1]	Normal	25.3	28.9	31.3	34.6	43.9	29.0	33.0	29.9	32.7	27.6	14.7	14.9	14.6	15.0	14.9
	Medium	25.0	27.4	29.8	33.3	41.5	27.5	30.7	25.3	24.8	16.4	15.0	15.0	14.7	15.0	15.0
	Low	24.9	24.8	30.6	32.3	39.8	25.8	28.5	21.0	19.0	10.7	14.8	14.6	15.1	15.0	14.9
N[1]	Normal	23.2	25.5	29.9	33.1	42.3	29.1	33.9	28.7	30.9	26.2	15.2	14.8	14.6	15.1	14.9
	Medium	23.4	25.0	26.9	32.6	40.5	32.2	34.1	27.4	29.6	24.4	8.9	8.7	9.0	9.6	9.3
	Low	23.2	23.3	25.4	28.1	35.6	32.9	34.5	24.4	25.8	23.0	4.3	4.3	4.4	4.6	4.6
Untreated		25.5	29.5	31.9	34.5	41.9	31.2	33.1	31.0	33.2	27.3	15.0	14.8	15.0	15.0	15.0
LSD (0.05)[1]		NS	3.1	3.0	2.9	4.0	3.1	3.6	3.7	3.3	2.8	0.5	0.4	0.3	0.4	0.5

[1] – Level of herbicide (H), moisture (M), and nutrient (N) stress.
[2] – Means separated according to Fisher's Protected LSD at $P = 0.05$.

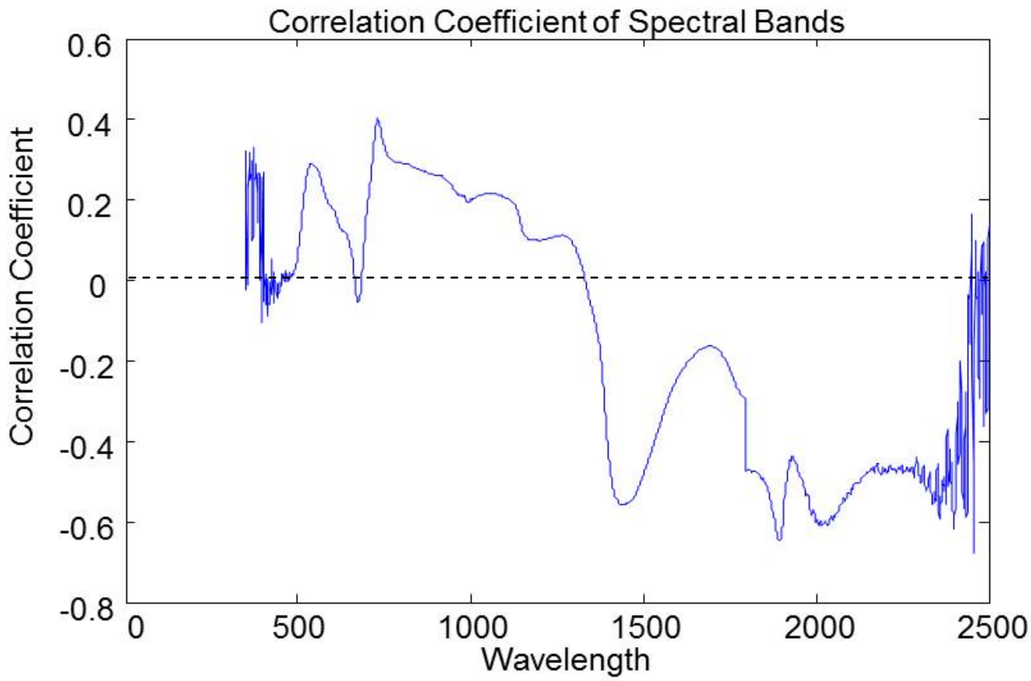

Figure 4.1. Spectral features recorded 18 DAE and 3 DAT which best correlate herbicide stress (positive correlation) vs. nutrient stress (negative correlation).

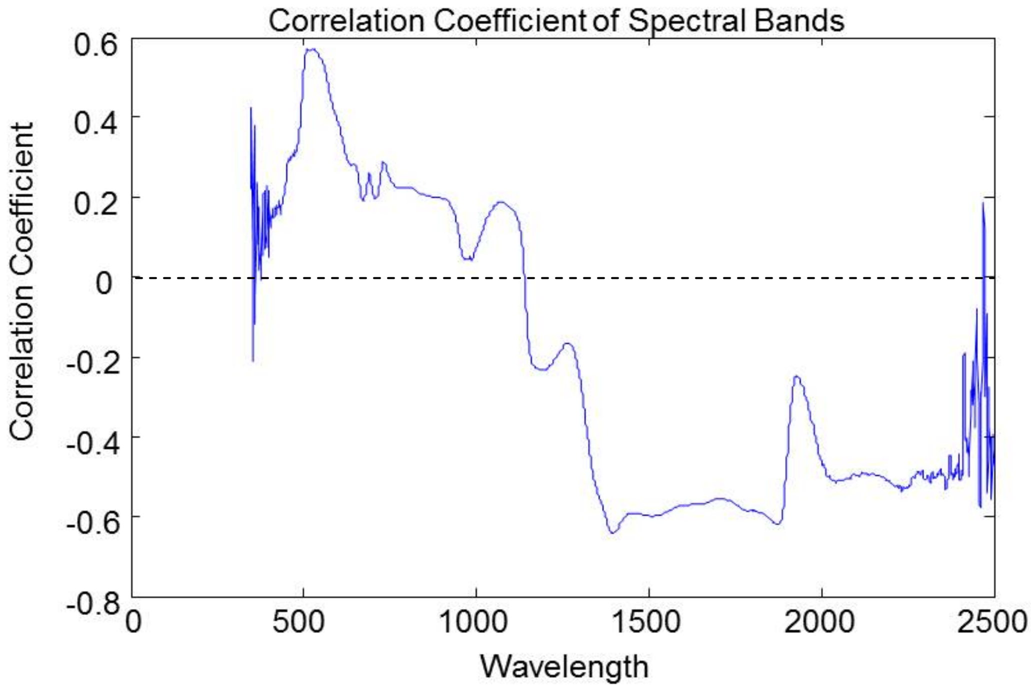

Figure 4.2. Spectral features recorded 22 DAE and 7 DAT which best correlate herbicide stress (positive correlation) vs. nutrient stress (negative correlation).

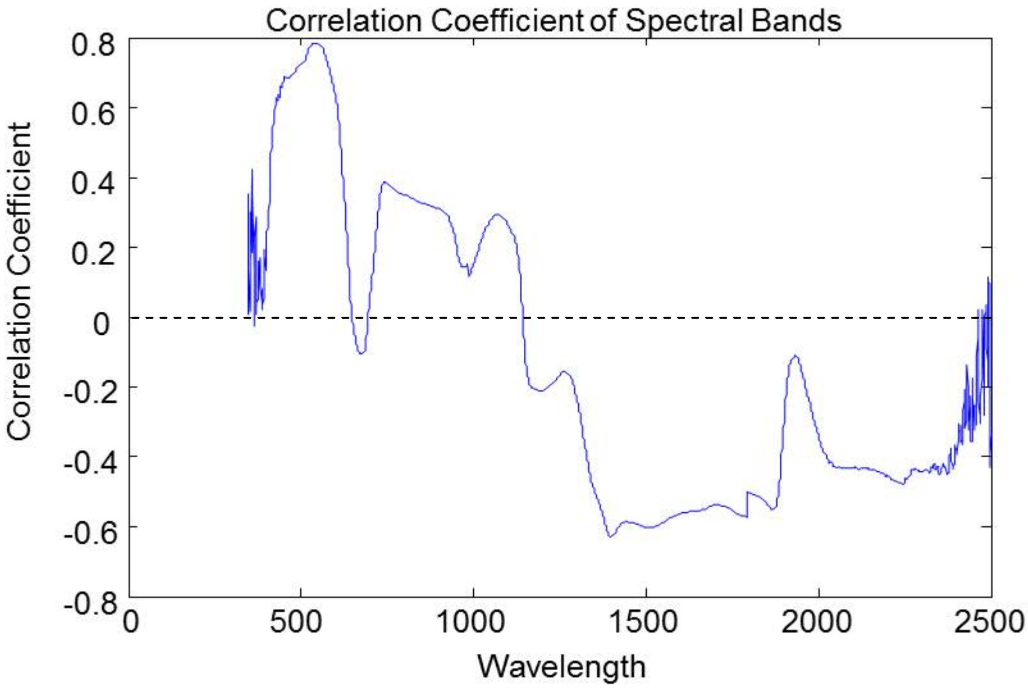

Figure 4.3. Spectral features recorded 29 DAE and 14 DAT which best correlate herbicide stress (negative correlation) vs. nutrient stress (positive correlation).

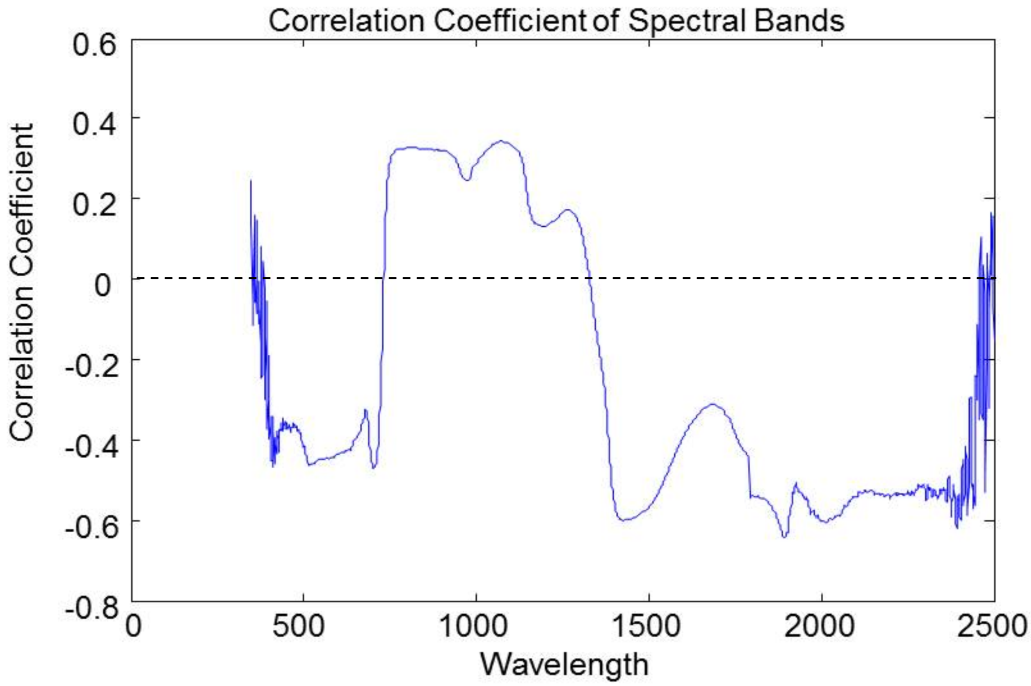

Figure 4.4. Spectral features recorded 18 DAE and 3 DAT which best correlate herbicide stress (positive correlation) vs. moisture stress (negative correlation).

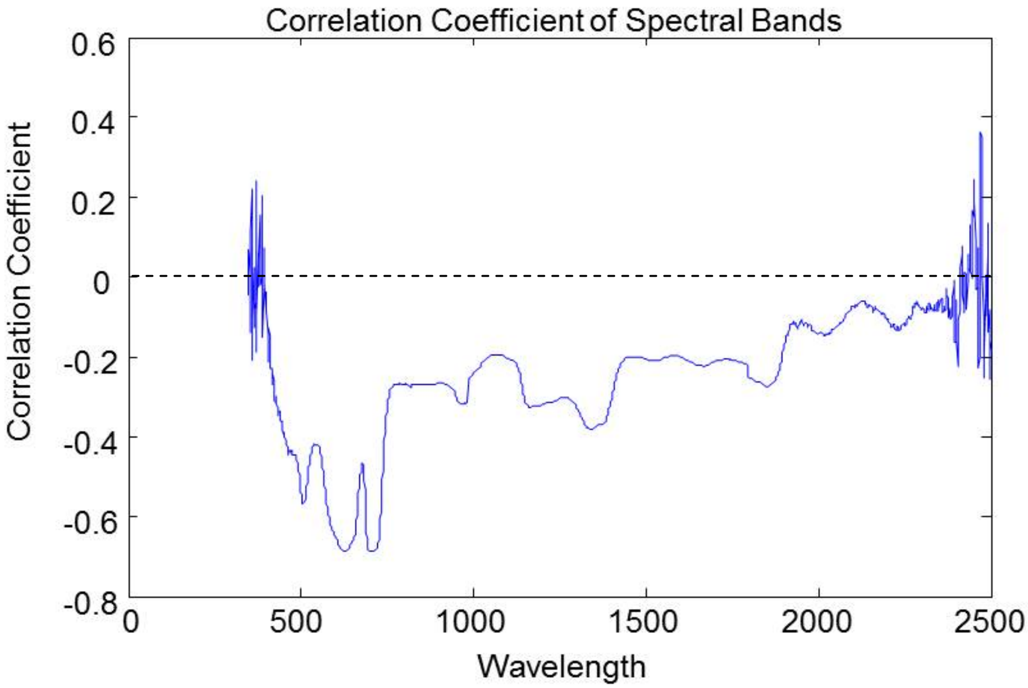

Figure 4.5. Spectral features recorded 22 DAE and 7 DAT which best correlate herbicide stress (positive correlation) vs. moisture stress (negative correlation).

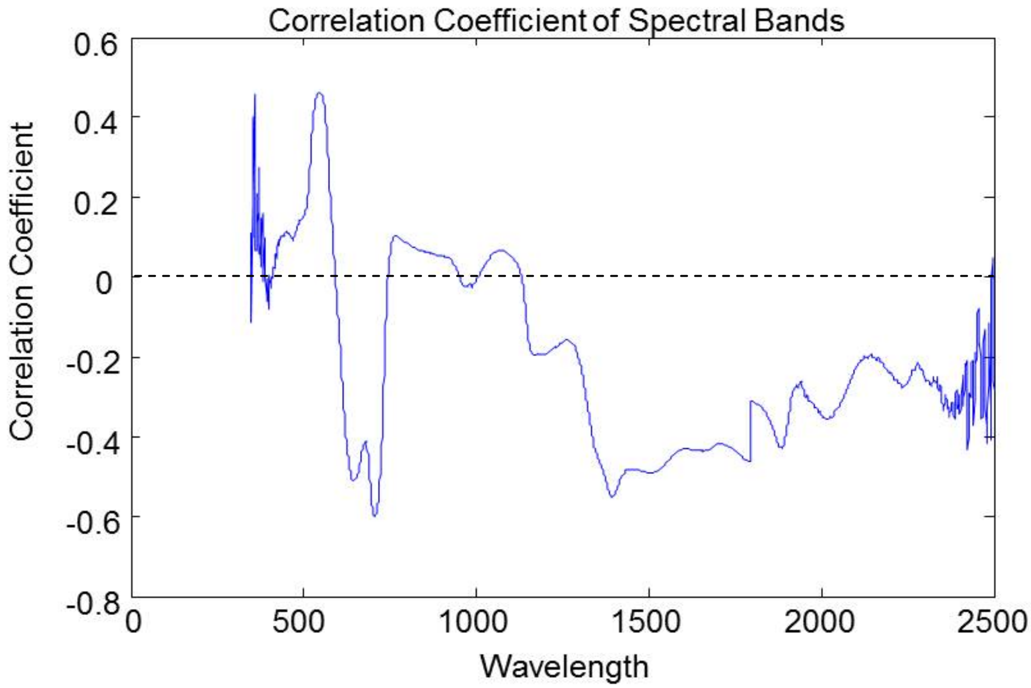

Figure 4.6. Spectral features recorded 29 DAE and 14 DAT which best correlate herbicide stress (positive correlation) vs. moisture stress (negative correlation).

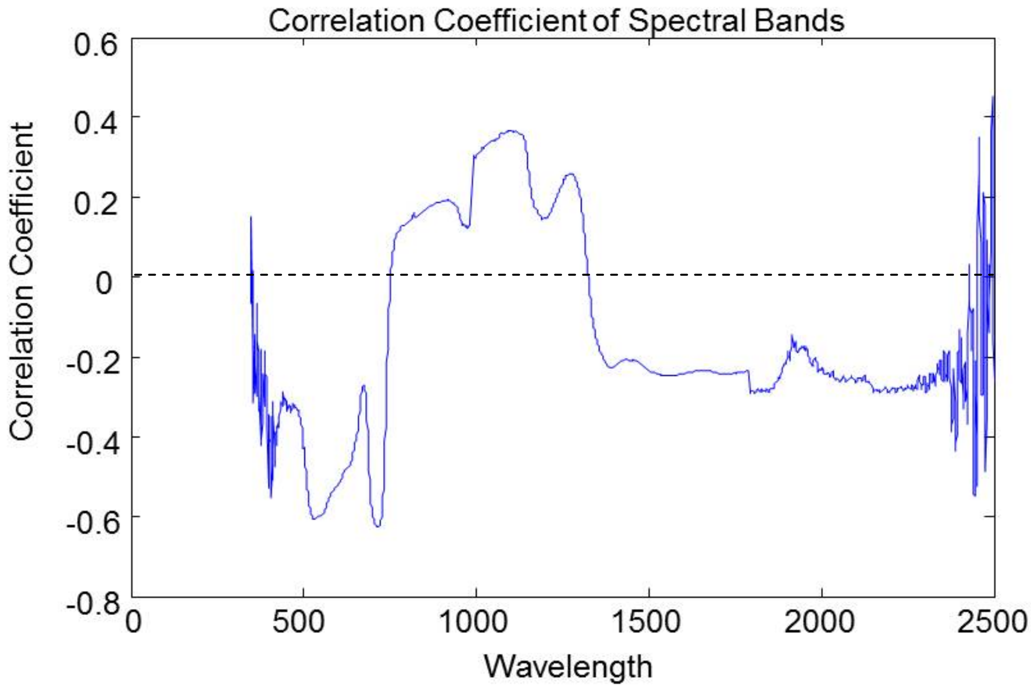

Figure 4.7. Spectral features recorded 18 DAE and 3 DAT which best correlate nutrient stress (positive correlation) vs. moisture stress (negative correlation).

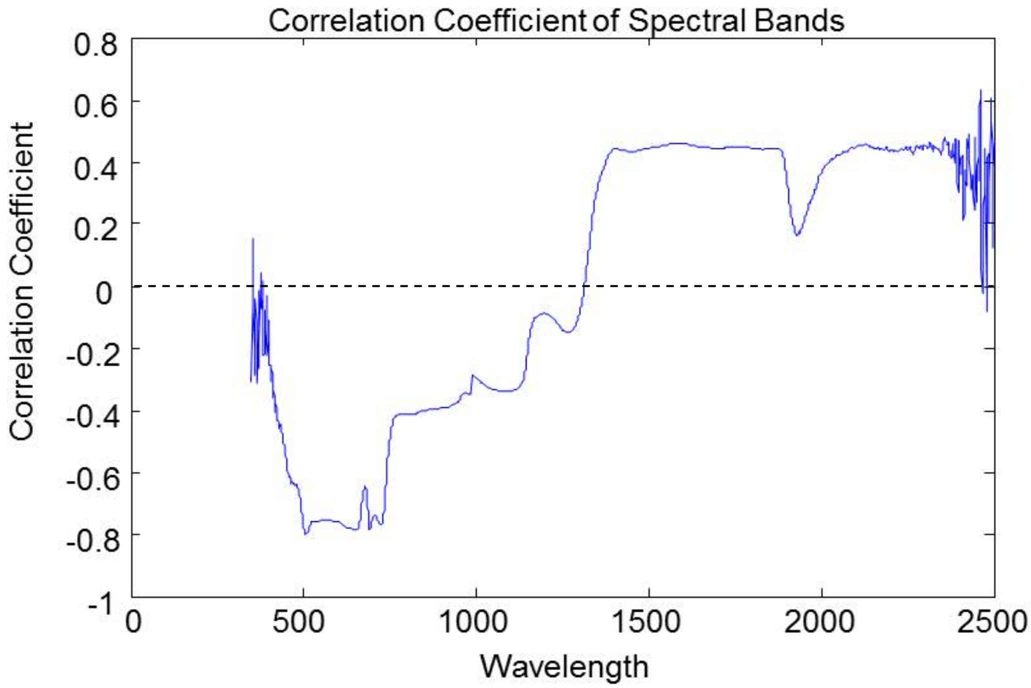

Figure 4.8. Spectral features recorded 22 DAE and 7 DAT which best correlate nutrient stress (positive correlation) vs. moisture stress (negative correlation).

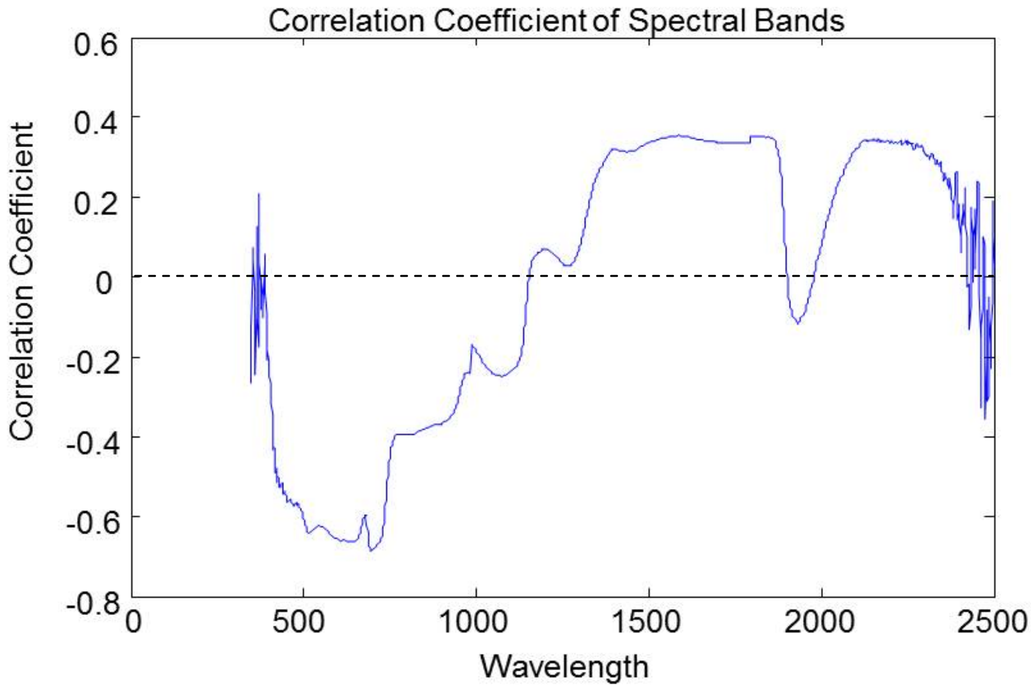

Figure 4.9. Spectral features recorded 29 DAE and 14 DAT which best correlate nutrient stress (positive correlation) vs. moisture stress (negative correlation).

Table 4.2. Spectral range within the electromagnetic spectrum which best describes herbicide, nutrient, or moisture stress 3 days after induced herbicide stress and 18 days after induced nutrient and moisture stresses.

	Spectral Range		
Stress Type[1]	Visible (400-700 nm)	NIR (700-1350 nm)	IR (1350-2500 nm)
Herbicide vs. Nutrient	Herbicide	Herbicide	Nutrient
Herbicide vs. Moisture	Moisture	Herbicide	Moisture
Nutrient vs. Moisture	Moisture	Nutrient	Moisture

[1] – Evaluation of spectral features associated with herbicide stress compared to nutrient stress, herbicide stress compared to moisture stress, or nutrient stress compared to moisture stress.

Table 4.3. Spectral range within the electromagnetic spectrum which best describes herbicide, nutrient, or moisture stress 7 days after induced herbicide stress and 22 days after induced nutrient and moisture stresses.

	Spectral Range		
Stress Type[1]	Visible (400-700 nm)	NIR (700-1350 nm)	IR (1350-2500 nm)
Herbicide vs. Nutrient	Herbicide	Herbicide	Nutrient
Herbicide vs. Moisture	Moisture	Moisture	Moisture
Nutrient vs. Moisture	Moisture	Moisture	Nutrient

[1] – Evaluation of spectral features associated with herbicide stress compared to nutrient stress, herbicide stress compared to moisture stress, or nutrient stress compared to moisture stress.

Table 4.4. Spectral range within the electromagnetic spectrum which best describes herbicide, nutrient, or moisture stress 14 days after induced herbicide stress and 29 days after induced nutrient and moisture stresses.

	Spectral Range		
Stress Type[1]	Visible (400-700 nm)	NIR (700-1350 nm)	IR (1350-2500 nm)
Herbicide vs. Nutrient	Nutrient	Nutrient	Herbicide
Herbicide vs. Moisture	Herbicide	Herbicide	Moisture
Nutrient vs. Moisture	Moisture	Moisture	Nutrient

[1] – Evaluation of spectral features associated with herbicide stress compared to nutrient stress, herbicide stress compared to moisture stress, or nutrient stress compared to moisture stress.

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