

ABSTRACT

Morgan, Sarah Paige. Estimating forage biomass and nitrogen concentration using false color infrared photography. (Under the direction of J.T. Green and J.G. White).

The objective of this research was to investigate the utility of using nonnormalized (raw) digital counts and vegetation indices (VIs) derived from false color infrared (FCIR) photography to estimate biomass (dry), nitrogen (N) concentration, and N uptake of several warm season forage canopies at several locations. In July 2000, FCIR aerial photography was obtained at an altitude of 854 meters from an experiment established in 1998 at the Caswell Farm in Kinston, NC to investigate realistic yield expectations (RYE) from warm season forages fertilized with swine (*Sus scrofa domestica*) effluent and ammonium nitrate (NH_4NO_3). The experiment consisted of three forage canopies (bermudagrass [*Cynodon dactylon* L. 'Coastal'], crabgrass [*Digitaria sanguinalis* L. 'Red River'], and volunteer warm season [80% native crabgrass, 20% forbs]) fertilized at five N rates (0, 224, 449, and 674 kg ha⁻¹ yr⁻¹) with either effluent or NH_4NO_3 in a stripped split plot design. Biomass, N concentration, and N uptake were measured and regressed against green (G [490 to 550 nm]), red (R [550 to 700 nm]), and near infrared (NIR [700 to 900 nm]) digital counts and seven VIs (NDVI, Green NDVI [GNDVI], DVI, RVI, Normalized NIR [NormNIR], Normalized Green [NG], and Normalized Red [NR]). There was an N source x N rate interaction for N uptake in bermudagrass (BG) and crabgrass (CG) canopies and for biomass and N concentration in BG. Differences due to N source (N source x VI) affected the

relationship between biomass and GNDVI in BG canopies and many of the relationships between crop response variables and VIs in VWS canopies. Biomass was best estimated by NIR digital counts in BG ($R^2 = 0.82$), NDVI in CG ($R^2 = 0.54$), and NormNIR in VWS ($R^2 = 0.86$). Nitrogen concentration was best estimated by NDVI in BG ($R^2 = 0.62$), NIR digital counts in CG ($R^2 = 0.56$), and G digital counts in VWS ($R^2 = 0.63$). Green NDVI was a consistently strong estimator ($R^2 > 0.76$) of N uptake for all forage canopies and was unaffected by N source.

In September, 2000, an experiment was established in Raleigh, NC to (i) test the utility of FCIR ground-based photography in estimating N concentration differences in bermudagrass ('Coastal') canopies grown at similar biomass, and (ii) to investigate how different soil moisture levels would affect image interpretation. The experiment consisted of three irrigation levels (0, 25 minutes, and 90 minutes) applied 24 hours before harvest split across three replications of five N rates (0, 11, 22, 45, and 90 kg ha⁻¹) applied eleven days before harvest on a well-established bermudagrass sod at early heading. False color infrared photographs were obtained from a height of 1.83 meters above the ground and represented a harvest area of 0.25 m². Biomass (dry), N concentration, N uptake, and soil moisture were measured at each harvest area and regressed against nonnormalized raw digital counts and VIs. Differences among N rates were found for N concentration and N uptake but not for biomass, indicating that biomass levels were similar across all treatments. Irrigation rates only affected soil moisture. Significant, but weak correlations ($R^2 < 0.28$) were found for the relationships among N concentration, N uptake, NIR digital counts, NormNIR, DVI, NG and GNDVI. When replications were analyzed as 'sites' and irrigation blocks as 'replications within sites',

there was a site x VI interaction for N concentration, NG, and GNDVI, whereby two of the three 'sites' were more strongly correlated ($R^2 = 0.38$ to 0.57) with a VI than the combined relationship. Relationships were generally stronger within site 1 versus site 2 and site 3.

In July and August of 2000 and 2001, ground-based FCIR photographs were acquired from six harvests of bermudagrass canopies ('Coastal') from four different locations throughout eastern North Carolina which were part of a larger experiment examining realistic yield expectations (RYE) on three soil types fertilized at five rates of nitrogen. Similar photographic methods were used, however, each harvest area consisted of an average of three photographs, each representing a ground area of 0.25 m^2 .

Relationships between Red, Green, and NIR digital counts and crop response variables for most of the sites were weak, however, normalization generally improved correlations. Moderate or strong correlations between spectral and crop response variables, such as between Green NDVI and N uptake ($R^2 = 0.89$), could be found among all sites and cuttings except one. There were cutting, year, and site interactions with VIs for all three comparisons among sites, cuttings, and years. Despite statistics indicating that harvests were best modeled individually, combined relationships usually resulted in higher coefficients of determination ($R^2 = 0.66$). Taking into account location, photography method, and environmental conditions, Green NDVI and DVI were best to estimate N concentration across four sites ($R^2 = 0.40$).

**Estimating Forage Biomass and Nitrogen Concentration
Using False Color Infrared Photography**

by
Sarah Paige Morgan

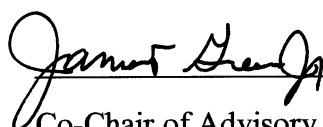
A thesis submitted to the Graduate Faculty of
North Carolina State University
in partial fulfillment of the
requirements for the Degree of
Master of Science

CROP SCIENCE

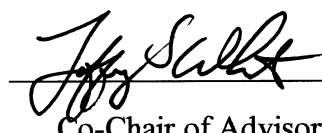
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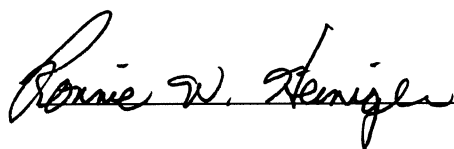
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BIOGRAPHY

Sarah Paige Morgan was born in Durham, North Carolina in 1976. She graduated from C.E. Jordan High School in 1994 with a strong interest in animal science and veterinary medicine. In August 1994, she began her undergraduate work in the department of animal science at North Carolina State University. She became active in the animal science club and developed a strong interest in livestock production. She worked and lived on the Butner Beef Cattle Research Farm in Butner, NC during her sophomore year and the Swine Educational Unit in Raleigh, NC during her junior year. These experiences provided her with an opportunity to work closely with livestock animals as well as participate in university research projects. During her senior year, she developed an interest in pastureland ecology and international travel while working as a research assistant for Dr. Matt Poore. This experience provided her the opportunity to participate in a research project examining weight gain of steers grazing stock piled fescue in a strip grazing design. While conducting this project Sarah met two individuals who would become critical to her success at NC State. Mike Scott, a graduate student, became an invaluable source of advice, support, knowledge, and humor and has continued to be a trusted friend and confidant. Finally, Dr. Jim Green, a forage extension specialist, became an inspiration for Sarah to pursue her interest in international travel and grazing management.

Sarah received her Bachelors of Science in Animal Science in May 1998. She was presented with a plane ticket as a graduation present from her mother and moved to

Matamata, New Zealand to work a season on a 600 head grass-based dairy farm. She spent ten months working for Steve and Trish Atkinson, whereby she gained a unique appreciation for intensively managing dairy cattle in a rotational grazing system. This experience also provided an opportunity for immense personal growth and inspired Sarah to continue her travels through Zimbabwe, Botswana, Namibia, and Australia. Sarah returned to North Carolina in August 1999 to begin her Masters Degree in the department of crop science with Dr. Jim Green.

ACKNOWLEDGEMENTS

I would like to thank Dr. Noah Ranells, Dr. Ronnie Heiniger for serving on my committee and supporting my research. I would like to thank Dr. Jeffrey White for serving as co-chair on my committee and providing critical technical and professional advice that was essential to the success of this research. I would like to acknowledge Dr. James T. Green Jr. for his constant support both financially and professionally, without which I would never have had the opportunity to participate in the International Grassland Congress in Brazil. No field research is possible without the help of competent and dedicated technicians. I would like to thank Michael Scott, Pete Thompson, and TJ Holliday for their field support, flexibility, and great attitudes while serving as members of my camera crew.

I would like to thank Dr. Gail Wilkerson, Mike Sturgill, and Greg Buol for their generosity in allowing me to take over a corner of their lab and providing computer technical support critical for keeping my research organized, manageable, and safe from viruses and crashes.

Graduate school can be a personally stressful journey and requires the support of friends and family. I would like to thank Dr. Jean-Marie Luginbuhl for all the coffees and stimulating conversation necessary to break up the amount of time I spent in front of the computer. I would like to thank my compadres, Cissy Elliott and Jennifer Tarter, for

being a constant source of inspiration and distraction, whichever was more appropriate at the time. A special thanks to my father, Craig Morgan, and my brother, Adam Morgan, for their continued interest in my scholastic evolution.

Finally, and most importantly, I would like to dedicate this thesis to my biggest fan, my mother. It has been my greatest fortune to have been challenged, comforted, encouraged, and unconditionally loved by a woman I admire and respect so much. None of my accomplishments would be possible without you and I thank you with all my being.

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INTRODUCTION

Groundwater contamination due to excessive nitrogen (N) fertilizers has become a serious problem in North America (Hubbard and Sheridan, 1989; Weil et al., 1990). North Carolina currently ranks number two among the United States in swine production and number one in broiler production, resulting in large amounts of nutrients being redistributed onto growing row crops and forages, primarily bermudagrass (*Cynodon dactylon* L.). Current methods to determine the efficiency of N fertilizers, such as cutting, drying, and analysis of forage samples, involve labor-intensive and costly sampling of plant tissue, soils, and animal manures. Due to the time consuming and variable sampling process, a rapid and noninvasive means of assessment would be beneficial

Remote assessment of N concentration and biomass from grassland canopies can provide land managers and regulatory agencies an opportunity to quickly determine the grassland inventory available for grazing animals, wildlife, or soil protection. False color infrared (FCIR) photography has been used to identify plant stress and yields for a variety of row crops, however its application for pasture-based grazing systems requires further study. This research project was designed to evaluate whether FCIR photography can be used as a reliable estimator of forage N concentration and biomass.

REVIEW OF LITERATURE

2.1 The Relationship Between Plant Growth and Reflectance

Solar radiation (500 to 2600 nm) that reaches the earth's surface may be absorbed, transmitted, scattered, or reflected by plant leaves (Gates et al., 1965). As a receptor, the human eye only perceives visible wavelengths (400 to 700 nm), while remote sensing instruments have the ability to measure reflected radiation beyond 5000 nm (Hatfield, 1990). The electromagnetic spectrum can be divided into three agronomically important regions: (a) visible light absorption (400 to 700 nm) which is dominated by pigments (chlorophyll a and b, carotene, and xanthophylls), (b) the near infrared (NIR) region (700 to 1300 nm) of high reflectance and low absorption affected most by internal leaf structure, and (c) the far infrared (IR) region (1300 to 2600 nm) which is most affected by the amount of water in the tissue (Thomas et al., 1967; Sinclair et al., 1971) (Figure 1). A plant leaf typically has low reflectance in the red and blue wavelengths due to strong absorption by chlorophylls, substantial reflectance in the green (the color we see), high reflectance in the NIR, and low reflectance in the IR wavebands (Figure 2).

2.1.1 Methods of Measuring Reflectance

Reflectance is defined as the ratio of the total radiant flux reflected by a surface to the total incident flux on the surface. Thus, only instruments that measure incident radiation (sunlight or internal light source) can calculate reflectance. Most early research on leaf spectral properties was done using spectrophotometers, which measure leaf absorbance of

a wavelength (500 to 2500 nm) by comparing the amount of incident radiation to the amount transmitted through a given sample (Thomas et al., 1967). Using two internal light sensors, chlorophyll meters approximate the amount of chlorophyll present in a leaf by measuring the amount of light that is transmitted through the leaf at 650 (Red [R]) and 940 (NIR) nm. Canopy radiation can be measured by elevating an instrument that measures reflectance over the canopy (e.g., a boom-mounted radiometer) or sensed photographically from ladders, planes, or satellites. Digital cameras and satellite imagery sense radiation from a canopy through a charge coupled device (CCD), converting the information into digital values.

Film-based sensing platforms do not measure incident light, rather they sense scattered and reflected radiation intensity from a given canopy and record it in three dye layers on film emulsions. False color infrared photography captures spectral radiation of vegetation in R, Green (G), and NIR wavelengths. In addition, all the layers are sensitive to B radiation, which is typically responsible for the degrading effect of haze in a photograph. A yellow filter is used over the camera lens to absorb B light before it reaches the film and limit the exposure of each layer to only one spectral region (Fritz, 1967). Green foliage is an efficient reflector of NIR radiation, which is reproduced as R on the false color image, while G radiation reproduces as B, and R radiation is reproduced as G (Fritz, 1967; Wallen et al., 1977). For example, trees that have been stressed will reflect less NIR radiation, making the trees in the photograph appear less R and more blue-green (Fritz, 1967).

2.1.2 Vegetation Indices

Spectral vegetation indices (VIs) are mathematical combinations of observations from one or more spectral wavelengths yielding a single quantity that may be more sensitive to plant responses than a single wavelength (Deering et al., 1975; Wanjura and Hatfield, 1987). An ideal VI would be highly sensitive to vegetation, insensitive to soil background changes, and only slightly influenced by atmospheric path radiance (Jackson et al., 1983). Reflectance in the NIR is often referenced to visible wavelengths, resulting in a ratio that should improve the sensitivity of the reflectance measurement to vegetation (Schepers et al., 1996) (Table 1). The Ratio Vegetation Index (RVI), first reported by Jordan (1969), has been used to normalize variations due to soil background reflectance, and generally forms a linear relationship with biomass (Colwell, 1973; Tucker, 1979). The Difference Vegetation Index (DVI) has shown similar correlations with biomass and chlorophyll content (Jordan, 1969; Tucker, 1979). The Normalized Difference Vegetation Index (NDVI) has been used more than any other VI in detecting and estimating plant responses, primarily N stress and biomass, and is a truly normalized index with values ranging from -1 to 1 (Deering et al. 1975, Tucker, 1979). Replacing the R band information with G resulted in the Green Normalized Difference Vegetation Index (GNDVI), which has been shown to be five times more sensitive than NDVI to canopies with high levels of chlorophyll (Gitelson, 1996). Jain (1989) suggested that individual wavebands could be normalized (e.g. Normalized NIR [NormNIR]) by dividing one waveband by the sum of the wavebands sensed. Each of these indices will be discussed throughout this review in context with the plant responses they measured.

2.1.3 Leaf Reflectance

Understanding the interaction between plant leaves and solar radiation was the basis for the development of canopy remote sensing applications. Early research examining the spectral properties of leaves resulted in the basic observation that plants absorb solar radiation efficiently where they require energy (visible wavelengths) and poorly in the NIR where the wavelengths are longer and have less energy (Gates et al., 1965). This relationship is primarily defined by plant pigments and internal leaf structure.

Visible wavelengths respond primarily to pigments located in the leaf mesophyll.

Chlorophyll is the most abundant of these pigments (65%) and has the most significant role in photosynthesis as a light-harvesting molecule (Gates et al., 1965). Kleshnin and Shul'gin (1959) reported that leaf pigments absorbed 70% to 90% of radiation in the blue (carotenoids) and red regions (chlorophyll a and b) of the visible spectrum, while absorbing the smallest amount (< 20 %) in the green, which was primarily reflected.

Gates et al., (1965) noted several trends in the spectra of a maple (*Quercus alba*) leaf as it aged. As chlorophyll content increased with the growth of the leaf, absorption increased in the R band (~680 nm) and decreased in the G and NIR wavelengths. When chlorophyll content reached a maximum value, the strong absorption bands (B and R) began to absorb in the G, slightly reducing the reflectance of the mature, dark leaf at 550 nm. As the leaf matured, the transition between R and NIR spectral response shifted more towards 700 nm. They concluded that spectral response in the visible wavelengths was related to the concentration of pigments in the leaf tissue. Knipling (1970) demonstrated that leaves lacking in the usual pigments reflected as much of the visible

light as they did in the NIR. These results were confirmed by Maas and Dunlap (1989), who found that in the absence of pigmentation in albino corn (*Zea mays*) seedlings, reflectance in the visible was the same as the maximum reflectance observed in the NIR. These results led to the conclusion that NIR reflectance was relatively insensitive to the presence of leaf pigments (Gates et al., 1965; Guyot, 1990).

Leaf reflectance measurements can be used to follow changes in leaf chlorophyll content which is related to N concentration in leaf tissue (Benedict and Swindler, 1961; Sinclair et al., 1971; Wolfe et al., 1988). Thomas and Oerther (1972) observed that limiting N in sweet pepper (*Capsicum annum* L.) leaves was accompanied by a reduction in chlorophyll concentration. Gausman et al. (1973a) reported a positive linear correlation between chlorophyll and G reflectance at 550 nm in corn leaves ($r^2 = 0.75$) for four stages of growth (seedling, pre-tassel, tassel and silk, and mature) and a negative correlation between chlorophyll and R reflectance at 650 nm in cotton leaves ($r^2 = 0.71$) for four stages of growth (seedling, pre-bloom, boll, and mature). Mature cotton leaves had higher NIR reflectance than younger leaves. In a study involving eight different crops (corn, cotton [*Gossypium hirsutum* L.], grain sorghum [*Sorghum bicolor* L.], tobacco [*Nicotiana tabacum* L.], cucumber [*Cucumis sativus* L.], lettuce [*Lactuca sativa* L.], spinach [*Spinacia oleracea* L.], and cantaloupe [*Cucumis melo* L.]), Thomas and Gausman (1977) reported an inverse relationship between chlorophyll and carotenoid concentrations and leaf reflectance at 450 (B), 550 (G), and 670 nm (R), however reflectance measurements at 550 nm were superior in relating leaf reflectance to chlorophyll concentration. These studies led to the conclusion that chlorophyll

concentration is inversely related to leaf reflectance in the R and B wavelengths and best detected by G wavelengths that include the 550 nm band.

Reflectance in the NIR is mostly a function of leaf structure, such as mesophyll structure or leaf thickness, and to a lesser extent leaf water content (Gates et al., 1965; Wooley, 1971; Sinclair et al., 1971). Near infrared wavelengths are scattered or reflected from leaf surfaces by refractive index discontinuities, the most important of which is the cell-wall/air-space interface (Gausman, 1974). In the maple leaf study, Gates et al. (1965) observed that NIR reflectance was highest for the leaf when the mesophyll consisted mostly of spongy parenchyma and lots of air spaces, which favor internal reflection. As the leaf matured, cells enlarged and the intercellular space decreased, resulting in a decrease in NIR reflectance. As leaves age, visible reflectance typically increases due to degradation of chlorophyll and other pigments, while NIR reflectance decreases due to deterioration of cell constituents (Knippling, 1970). Senescence causes changes in leaf pigmentation and structure, and can occur naturally as canopies mature or are stressed by lack of water and nutrients (Sanger, 1972). Gausman (1974) investigated the effects of air space on leaf reflectance by replacing the air spaces in cotton leaves with castor oil and water, both of which resulted in decreased reflectance between 500 and 2500 nm. In the same study, Gausman (1974) noted that stressed leaves usually had lower reflectance than nonstressed leaves if the leaves were the same age. However in a growing crop, stressed leaves of the same age could be stunted, yellowing (nutrient deficiency), or dehydrated, which would result in a higher NIR reflectance. Younger leaves had more compact cell structure (low NIR reflectance / high transmittance), while aged leaves were

more porous (high NIR reflectance / low transmittance). Wooley (1971) observed increased reflectance in the visible and NIR portions of the spectrum when corn leaves were water stressed. Dried leaves had higher reflectance than fresh green leaves for corn and soybean (*Glycine max*). He concluded that leaf thickness had more influence on transmittance than reflectance. Temperature extremes can result in increased reflectance. Wooley (1971) noted that frost or freezing conditions can result in increased reflectance due to the refractive index differences that occur with the crystallization of water, and that subsequent thawing can result in lower canopy reflectance due to water flowing into the intercellular spaces of the leaf tissue. Mitchell et al. (1990) observed increased R reflectance from frost-injured alfalfa pastures.

Mesophyll structure and reflectance properties differ among species. A corn leaf has a compact mesophyll and will have lower reflectance and higher transmittance as compared with leaves with more porous mesophyll, such as maples leaves (Gausman, 1974). In a study of six agronomic crops (corn, sorghum, soybean, wheat [*Triticum vulgare* L.], oat [*Avena sativa* L.], and sudangrass [*Sorghum sudanense* (Piper)], Sinclair et al. (1971) observed similar leaf reflectance values obtained from a spectrophotometer (500 to 2,600 nm) for fresh turgid green leaves for all species. However, higher reflectances were noticed for corn (monocot) leaves versus soybean (dicot) leaves sampled at maturation and senescence.

Some scientists have observed saturation effects in the visible and NIR. In 1961, Shul'gin et al. identified a chlorophyll content threshold of 3 mg 100 cm⁻², whereby light

absorption changes occurred primarily below this threshold. Sanger (1972) found that chlorophyll concentration decreased rapidly as plants matured and biomass plateaued. These observations indicated that when a leaf reached maximum chlorophyll content and growth continued, visible reflectance was not a sensitive indicator of yield. Conversely, when leaves reached maximum growth and chlorophyll concentrations started to decrease, NIR reflectance was not a sensitive indicator of N concentration. Increased NIR reflectance and decreasing R reflectance have been noticed among individual leaves with increasing leaf area index (LAI), percent vegetation coverage (Gausman et al., 1976), N fertilization (Walburg et al., 1982), and biomass (Colwell, 1974). The opposite trend has been associated with plant stress (Olson, 1969) and changes in leaf geometry (Colwell, 1974). Hinzman et al. (1986) found that leaf reflectance of wheat in the visible wavelengths was lower than bare soil reflectance, but higher than bare soil in the NIR wavelengths. They further noticed that as wheat leaves matured and senesced, their reflectance measurements approached those of bare soil. Gausman et al. (1976) identified maximum reflectance at an LAI of two in the visible wavelengths, and eight in the NIR wavelengths for corn leaves 48 days after emergence.

Vesk et al. (1966) found that N treatment differences resulted in changes in leaf structure, composition, pigment concentration, cell size, and cell wall composition and structure. Therefore, changes in N availability will usually cause changes in leaf reflectance. Several studies analyzing spectral properties of individual leaves from a variety of crops have shown that reflectance in the visible spectrum increases as N becomes more deficient, indicating that limiting N reduces the concentration of chlorophyll, and that

chlorophyll is an efficient absorber of light (Thomas and Oerther, 1972; Al-Abbas et al., 1974; Wanjura and Hatfield, 1987; Hinzman et al., 1986; Takebe et al., 1990; Blackmer et al., 1994). Reflectance measurements in the visible wavelengths can indicate levels of plant stress because chlorophyll absorption is highly sensitive to metabolic disruptions, such as N deficiency or drought (Knippling, 1970). Olson (1969) found that leaves of various tree species under N stress decreased in NIR reflectance and increased in R reflectance when compared with leaves not under N stress. Thomas and Oerther (1972) found that leaf reflectance measured at 550 (G) and 675 (R) nm could be used to estimate the N status of sweet pepper leaves. Al-Abbas et al. (1974) detected decreases in chlorophyll content resulting from nutrient stresses in corn by comparing leaf reflectance measurements at 640 (R) and 530 (G) nm. Differences in N concentrations for greenhouse-grown soybean plants six weeks after germination were best detected when examining leaf reflectance measurements at 550 (G) nm (Chapelle et al., 1992). Blackmer et al., (1994) found individual leaf reflectance near 550 (G) and 710 (NIR) nm to be strongly related to N stress in corn.

2.2 Factors Affecting Canopy Reflectance

Canopy components such as canopy architecture, nutrient stress, soil background reflectance, and atmospheric effects can cause a canopy to reflect less visible and NIR radiation than an individual leaf and thus must be considered in remote sensing of plant canopies in the field (Daughtry et al., 1980; Hatfield, 1990). Knippling (1970) noted that this difference was greater for visible reflectance than NIR reflectance sensed from a

vertical angle. This can be credited to the transmission of wavelengths from the upper leaf layers in a canopy to the lower leaf layers and soil where radiation is scattered, resulting in a darker shadow effect in visible wavelengths versus NIR wavelengths (Curran, 1983; Campbell, 1996). Data obtained spectrophotometrically and photographically show that much of the NIR light transmitted through the uppermost leaf layers is reflected from the lower leaves and retransmitted up through the canopy resulting in an enhanced reflectance (Myers et al., 1966). In visible wavelengths, the first leaf layer of a canopy, if healthy and perpendicular to the incident light, absorbs approximately 90% of the incident light (Gausman et al., 1973b). In NIR wavelengths, the first leaf layer absorbs about 10% of the incident light, the rest being about equally divided between transmission and reflection (Monteith, 1965). Tucker (1977) reported that NIR radiation transmitted by upper leaf layers is scattered by lower leaves in the canopy until the incident light is attenuated by a LAI of eight for various grass. Red reflectance from a canopy may only represent the uppermost leaf layers due to the intense absorption by chlorophyll, while NIR reflectance (low absorbance) may represent multiple leaf layers in the canopy (Heute, 1987).

2.2.1 Canopy Architecture

Leaf Area Index. In general, visible reflectance in a canopy is negatively associated with LAI, percent vegetative cover, and green biomass, while NIR reflectance is positively associated with these canopy parameters. Vegetation cover (area of ground covered by green leaves) is the single canopy characteristic that has the most influence on R and NIR reflectance (Curran, 1983). Leaf area index, the total one-sided leaf area

measured over a horizontal unit of ground area, is closely related to the proportion of green vegetation in a canopy (Carlson and Ripley, 1977). While being a good representation of canopy biomass, LAI is often difficult to quantify with spectral measurements due to its three dimensional nature. Sensitivity of spectral measurements to LAI weakens and reaches a threshold as LAI increases from 2 to 4 (depending on vegetation type), reducing their utility under conditions of dense vegetation cover (Carlson and Ripley, 1977; Curran, 1983).

Several scientists have observed a lack of sensitivity of R reflectance in canopies where LAIs were greater than three, indicating that the canopy had reached a chlorophyll threshold (Shul'gin et al., 1961; Tucker, 1979; Kollenkark et al., 1982; Hinzman et al., 1986). Walburg et al. (1982) found that RVI was a more sensitive indicator of LAI of a corn canopy ($r^2 = 0.77$) than either NIR or R reflectance over two growing seasons, but approached an asymptote at LAI values above three. Hatfield et al. (1983) found similar linear relationships between RVI and LAI of wheat canopies over various planting dates. Tucker (1979) noticed similar trends for NDVI at LAI > 3. Asrar et al. (1984) followed changes in NDVI of a wheat canopy sensed with a radiometer throughout the growing season. When LAI was zero early in the growing season, NDVI was equal to the ratio of soil reflectance in the R and NIR. Values for NDVI increased as LAI increased with canopy growth, reaching a plateau at LAIs greater than six. As LAI decreased with senescence, NDVI values also decreased. Linear plateau relationships between NDVI and LAI have been reported for a variety of crops (Tucker, 1979; Holben et al., 1980 [soybean]; Best and Harlan, 1985 [oat]; Gallo et al., 1985 [corn]; Sellers, 1985).

Stage of Growth. Reflectivities of plant canopies change as they mature and accumulate biomass. During early vegetative growth of most row crops, canopy cover is low and reflectance is influenced primarily by soil background. Ashley and Rea (1975) found NDVI ratios to improve with foliar development of a forest canopy and decrease with senescence. Wanjura and Hatfield (1987) conducted an extensive study analyzing four crops (cotton, soybean, grain sorghum, and sunflower [*Helianthus annuus* L.]) at two stages of growth (vegetation and maturation) using a radiometer (height above ground [HAG] = 4.3 m) that measured two wavebands (NIR = 760 to 900 nm; R = 630 to 690 nm). They regressed RVI and NDVI against fresh weight, dry biomass, LAI, and percent ground cover. Correlations for RVI and NDVI with LAI were consistently higher during the vegetative phase for all crops, however there were greater variations between the two growth stages for cotton and sunflower. Dry biomass was correlated ($r^2 > 0.60$) with RVI and NDVI for all crops in the vegetative phase. They concluded that RVI should be used to estimate LAI and biomass when a crop is near peak vegetative growth but during early vegetative growth NDVI is a better estimator of LAI and biomass. Wiegand et al. (1986) suggested that spectral differences in crops during the maturation phase may be due to different rates of senescence and subsequently different amounts of nonphotosynthetically active leaves. Jackson et al. (1983) found RVI to be most sensitive at > 50% canopy cover for wheat under two irrigation treatments.

Near infrared reflectance is less sensitive to senescing leaves in a canopy than visible canopy reflectance. Colwell (1974) measured reflectance of all-green and all-dead oat

canopies under equivalent percent covers and environmental conditions. Leaf reflectance in the NIR increased by 10 % when sensing from green to dead leaves, while NIR canopy reflectance increased only 3 %. For the same canopy, R reflectance of the leaves increased by 50% from live to dead, while canopy reflectance increased 33%. His observations indicate that R radiation is more sensitive to differences in chlorophyll than NIR radiation, if other canopy characteristics are similar.

Leaf Geometry. The arrangement of leaves on the stem and orientation to the sun (leaf orientation) has a much greater impact on measurements sensed from the canopy than those sensed on individual leaves (Hatfield, 1990). Plants that exhibit horizontal leaf growth will have a greater concentration of palisade cells toward the upper surface of the leaf or the side that receives the most light, while leaves with more vertical growth patterns will have equal amounts of palisade cells on both sides of the leaf (Wooley, 1971). Suits (1972) devised an analytical model of vegetation canopy reflectance to show that NIR reflectance may decrease and R reflectance may increase when some of the leaves change from a predominantly horizontal to vertical orientation. Using similar canopy modeling techniques, Colwell (1973) showed that decreases in LAI can also cause NIR canopy reflectance to decrease and R reflectance to increase without any change in individual leaf reflectance. Pinter et al. (1985) observed that wheat canopies with more horizontal leaves exhibited higher reflectance and less sensitivity to solar zenith angle in the visible and NIR regions of the spectrum than canopies with more vertical leaves.

Wooley (1971) noticed differences in canopy reflectance for corn and soybean plots when lodging conditions resulted in a mixture of fronts (adaxial) and backs (abaxial) of leaves facing a spectrophotometer. Typically, the palisade tissue in the adaxial side contains more chloroplasts and less intercellular air space, while the abaxial side is composed of less densely packed chloroplasts and three to four times more intercellular air space. He observed greater reflectance in the NIR region when the adaxial side was facing the sensor. Colwell (1974) noticed a 50% increase in R reflectance when he "smoothed out with several strokes of the hand" a lodged grass canopy at 100% vegetative coverage. He concluded that shadows caused by lodging resulted in more variation in the R spectral region than in the NIR, and that RVI would not be entirely effective in normalizing variations due to shadow or altered leaf angle. Additional studies have found that lodging conditions in a canopy can cause unusually high reflectance in the visible, particularly R reflectance, and the NIR (Stanhill et al., 1972; Colwell, 1974; Hinzman et al., 1986).

The amount of shadow in a vegetation canopy can affect canopy reflectance (Colwell, 1974). Reifsnyder and Lull (1965) found significant differences in NIR reflectance in shaded areas of a coniferous versus a hardwood forest. They suggested that thicker leaves in the coniferous canopy contributed to a darker shading effect and lower transmittance of light. Vinogradov (1969) attributed increased shade from vegetative cover to a negative correlation between reflectance and percent cover in the visible spectrum. Gerberman et al. (1976) observed a shadowing effect from upper leaves in cotton canopies sensed aerially, thus reducing reflectance and giving the impression of

less than 100% ground cover. Quality of light is altered when light is transmitted from upper leaves of a canopy to the lower leaves. Weather conditions, such as wind, rain, and temperature, can also alter leaf orientation enough to affect canopy reflectance.

Cultivar. Crop cultivars that differ in maturation rates, leaf orientation, and pigmentation can also differ in spectral response. Significant variety differences in canopy radiation sensed aerially have been observed in corn and wheat (Leamer et al., 1978; Blackmer and Schepers, 1996; Flowers et al., 2001; Hatfield, 1990). Kollenkark et al. (1982) attributed early season spectral differences among soybean cultivars to differences in soil cover, with one cultivar reaching canopy closure before the other two cultivars. Later in the season, each cultivar began senescing at different times and rates, resulting in significant differences in NIR reflectance. Stone et al., (1996) found effects of variety and variety by N rate on relationships between total N uptake and the Plant Nitrogen Spectral Index (PNSI), an inverse calculation of NDVI, on wheat. Similar results were found by Flowers et al. (submitted) who reported that wheat variety affected the relationship between all spectral indices tested (NIR, NormNIR, NDVI, RVI, DVI) and tiller density at GS-25. They observed differences in color and leaf orientation among varieties. Within-field tiller density references for each wheat variety were needed in both cases to improve relationships.

Weeds, especially broad-leaved species, have similar effects as cultivar on spectral reflectance of crops. Flower et al. (submitted) reported an improvement in the relationship between tiller density and NIR digital counts, when plots infected with

italian ryegrass (*Lolium multiflorum*) were removed from the data. In the same study, they also noticed decreased spectral values for all indices in plots where chickweed (*Stellaria media*) and henbit (*Lamium amplexicaule*), both broadleaf weeds, were present. These observations provide additional challenges to remote sensing of pasture systems where forage mixtures can consist of warm and cool season grasses and legumes.

2.2.2 Background Reflectance

Soil Reflectance. Background reflectance can confound canopy reflectance at low levels of vegetation cover (Daughtry et al., 1982). Heute (1987) observed that RVI ratios derived from similar vegetative cover on dark-colored soil were higher than those derived from light-colored soils, and closely approached RVI ratios for full canopy coverage. He concluded that soil reflectance contributed more to the overall reflectance in the NIR wavebands than the visible wavebands since canopies transmit more light in this region of the spectrum than they absorb, and thus allow more opportunity for soil to interact with the canopy reflectance. Elvidge and Lyon (1985) reported that NDVI overpredicted biomass when vegetation was incomplete and soils were dark. Kollenkark et al., (1982) observed significantly lower R and NIR reflectances, obtained from a radiometer (HAG = 3.4 m), of a soybean canopy on a dark-colored soil versus a light-colored soil until the canopies reached 80% vegetation cover. In seasonal growth studies of wheat, the variation in R and NIR reflectance when soils were visible was compounded in most VIs (Broge and Mortensen, 2002).

Soil Moisture. Drying of the soil surface has been associated with increasing spectral reflectance (Irons et al., 1989). Daughtry et al., (1980) identified soil moisture as one of the primary agronomic factors affecting reflectance of spring wheat from tillering to maturity in semi-arid environments. Soil moisture accounted for 73% of variation in R reflectance and 69% of the variation in NIR reflectance. Stoner et al., (1980) noted that increasing soil moisture decreased reflectance in all wavelengths. Some scientists have found it difficult to distinguish between soil reflectance and vegetation of some grassland species in arid environments, particularly when portions of the canopy are senescing, resulting in an inability to make accurate biomass estimations (Elvidge and Lyon, 1985; Heute et al., 1984). Precipitation on or before the day of sensing can cause canopy reflectance to decrease, especially when the additional moisture results in a change of soil color (Daughtry et al., 1980; Hinzman et al., 1986), can mask the effects of bare soil when canopy cover is low (Walburg et al., 1982). Irrigated soils exhibit irregular drying patterns, which often translate into different soil colors, making spectral responses associated with vegetation-related parameters (e.g. biomass, LAI, stress, disease) difficult to detect (Heute, 1987). Thompson (1976) noticed similar decreases in canopy radiance following precipitation using Landsat Multispectral Scanner (MSS) imagery, and was able to delineate precipitation patterns and drought severity. Mayhew et al. (1984) reported no effect of moisture on RVI readings from a mixed sward canopy sensed after a rain (HAG = 1.25 m).

Leaf Litter. Leaf litter (dead leaves) or thatch can have similar effects as soil background on canopy reflectance. Some studies have found that relatively small

increases in standing dead vegetation resulted in large effects on overall reflectance.

Heute and Jackson (1987) used a first order canopy model to show that litter and senescent leaves mixed with green vegetation in an arid grass rangeland resulted in decreased NDVI which was unable to detect small amounts of green vegetation.

Combining yellowing and senescing plants with bare soil resulted in an increase in NDVI. Sellers (1985) reported that dead material in a canopy decreased NDVI when the canopy LAI was less than six.

Vegetation indices are particularly useful in normalizing effects of background reflectance. For example, Jackson et al. (1983) observed decreases in NIR reflectance for wheat plots at jointing for fifteen days after irrigation due to a darkening of the soil.

When comparing these results with stress-induced wheat plots at the same growth stage, NIR reflectance appeared higher and erroneously indicated a greater amount of green vegetation for the stressed plants. They found RVI to be much less influenced by changes in soil reflectance caused by soil water content changes. The NDVI was more sensitive to soil background reflectance than RVI, but was able to detect vegetation at 15% cover versus 50% cover for RVI. The sensitivity to green vegetation of NDVI plateaued at 80% cover, while DVI decreased as IR radiation got higher and R radiation got lower. Colwell (1974) and Pearson et al. (1976) observed insensitivity of RVI to vegetation at canopy covers below 30%.

While soil background reflectance can be highly variable depending on degree of canopy coverage, soil type, and soil color, it is also predictable (Curran, 1983; Satterwhite and

Henley, 1987). Tucker and Miller (1977) reported high spectral contrasts between R and NIR reflectance of dry soil and dry biomass. Heute (1988) developed the Soil Adjusted Vegetation Index (SAVI) to minimize the influence of soil on vegetation reflectance by taking into account soil type and LAI. Under conditions of less than 100% canopy cover, the SAVI has been found to be superior to NDVI in predicting green vegetation (Goel and Qin, 1994; Leprieur et al., 1994). Variability due to soil background reflectance often requires photographic responses to be calibrated within fields (Colwell, 1974). Grain crops evaluated at early stages of growth often have bare soil effects due to row spacing.

2.2.3 Nutrient Stress

Nitrogen deficiencies have been associated with decreasing amounts of chlorophyll (Wolfe et al., 1988), which can translate into an increase in light reflectance in the visible wavelengths (Hinzman et al., 1986 [wheat]; Takebe et al., 1990 [rice]; Blackmer et al., 1994 [corn]). Walburg et al. (1982) reported an increase in R reflectance from N-deprived corn canopies, while NIR reflectance decreased. Blackmer et al. (1994) concluded that light reflectance near 550 nm (G) could be used to detect N deficiencies in corn. Stone et al. (1996) demonstrated that total plant N in wheat could be estimated using spectral radiance measurements from a sensor at 671 (R) and 780 (NIR) nm with enough accuracy to allow for in-season corrections of N deficiency. Hagger et al. (1984) used the same wavelengths measured by a hand held meter to discriminate between white clover and N-deficient grasses.

Chlorophyll meters are most sensitive when N nutrition is adequate or below, and have difficulty assessing excess N availability (Cerrato and Blackmer, 1991; Schepers et al., 1992). Wood et al. (1992) found a high correlation between field chlorophyll meter readings and corn tissue N concentration at the V10 (tassel begins to develop, stalk is elongating rapidly) and mid silk growth stages. Schepers et al. (1992) observed that chlorophyll meter readings of corn canopies increased with N rates but plateaued when N was adequate, making luxury N concentration difficult to estimate. Once N supply was adequate, the visible reflectance of corn canopies changed very little with increasing N rates (Blackmer et al., 1996b). Individual readings from chlorophyll meters can vary up to 15% from plant to plant and are not the best estimate of overall canopy N status (Peterson et al., 1993).

Nitrogen deficiencies can be detected by calibrating photographic responses within fields against responses from areas measured as sufficient in N (Blackmer et al., 1996a). Using digitized color slides derived from aerial photographs (HAG = 1000 m) of corn canopies just before harvest, Blackmer et al. (1996a) found that R relative digital counts (digital count of a primary color divided by the mean digital count of the reference N rate plot) provided better detection of N deficiency than the G or B relative digital counts.

2.3 Factors that Effect the Quality of Remote Sensing

2.3.1 Light Conditions

Quality of light conditions can have significant effects on reflectance values and spectral ratios. Milton (1981) suggested that the best method for controlling ambient light was to obtain spectral information under uniform irradiation conditions. Mayhew et al. (1984) reported that variation due to cloud cover significantly increased spectrophotometer R / NIR ratio readings of a mixed grass sward. The most consistent light conditions found for their study in southwest Scotland was under heavy cloud cover, when they observed no effect of solar angle between the hours of 0900 and 1600 in winter. Many researchers have found that spectral data is best collected on cloud-free days between the 1100 and 1300 hours to minimize the effects of shading on vegetation (Duggin and Phillipson, 1982).

Different solar angles can cause significant changes in visible and NIR reflectance as well as VIs derived from them (Pinter et al., 1983). Using an analytical canopy model, Colwell (1974) observed that as solar zenith angle decreased from 50 degrees to 10 degrees, R reflectance sensitivity to changes in vegetative cover also decreased. Pinter et al. (1983) found that larger solar zenith angles had a better relationship with wheat canopies with small LAIs. Hinzman et al., (1986) noticed some variation in data taken over a two-hour period with a spectrophotometer (HAG = 6 m), which they attributed to changes in solar azimuth and zenith angles during the sensing period. Vickery et al. (1980) noticed that reflectance at all wavelengths declined linearly with increasing zenith

angle. To compensate for increasing radiation with sun angle, data can be normalized to a standard sun elevation of 45 degrees (Pinter et al., 1983). If experimental plots are sensed individually, it can take much longer to collect data from an entire experiment, resulting in a range of solar angles. This can create a source of variability that is not easily quantified. If certain temporal data is recorded (date, time of day, longitude, and latitude) it is theoretically possible to make corrections based on changing solar angles.

2.3.2 Height Above Ground

Several researchers have found that reflectance measurements are more affected by atmospheric conditions the higher above the canopy they are obtained (Gerbermann et al., 1976; Scharf and Lory, 2000). Satellite imagery can be obtained from 161 (LANDSAT) to 681 km (IKONOS) depending upon the orbit. Spectral data obtained from satellites can be filtered into narrow bands and have been significantly correlated with LAI, green biomass, percent ground covered by vegetation, and chlorophyll concentration (Wiegand et al., 1971; Rouse et al., 1973; Deering et al., 1975; Richardson et al., 1975; Richardson and Wiegand, 1977; Thomas and Gausman, 1977). Pollock and Kanemasu (1979) identified the scattering and absorption of various particles between the satellite and the canopy as the primary limitations to using satellite data. Atmospheric path radiance effects can contribute to falsely large ratio values in satellite-based imagery (Dave, 1980; Switzer et al., 1981). The RVI and NDVI measurements can be severely depressed by changes in atmospheric radiance, being reduced by as much as 50% when sensing through a clear compared to a turbid atmosphere (Jackson et al., 1983). When sensing green vegetation, R reflectance (denominator for RVI) will be small, amplifying

atmospheric effects. Jackson et al. (1983) developed the Difference Difference Index $((2 * MSS7 - MSS6) - (MSS5 - MSS4))$ which reduced the effect of atmospheric scattering to less than 5%, however, it was particularly affected by soil reflectance. They concluded that this index could be a sensitive indicator of early vegetation if soils were dark and exhibited little change in reflectance with water content changes. Holben (1986) noted that scattering in the atmosphere by aerosol particles and molecules increases R reflectance substantially and thus decreases NDVI values. Large differences have been observed between NDVI measurements derived from satellite imagery and those measured at the canopy surface (Carlson and Ripley, 1997). Variations in soil brightness have been identified as the cause of variations in NDVI values from one image to the next (Liu and Heute, 1995), and accurate atmospheric corrections must be made to band ratios derived from satellite imagery in order for results to be reliable. Numerous mathematical formulas have been derived in recent years to account for seasonal and global atmospheric characteristics.

At altitudes ranging from 100 to 1500 meters, FCIR aerial photography has been reported to detect a variety of crop characteristics for many plant species (Blackmer and Schepers, 1996; Fent, 1999; Flowers et al., 2001). Gerbermann et al. (1976) found that ground cover could be estimated within 10% from low altitude (640 to 1219 m) FCIR aerial photographs as compared to within field visual estimates for various row crops. They obtained strong correlations between ground measurements and predictions based on aerial photographs for cotton ($r^2 = 0.98$) and grain sorghum ($r^2 = 0.95$). However, there was a tendency to over-estimate ground cover with aerial photographs for cotton, corn,

grain sorghum, and forage sorghum due to incomplete canopy coverage and shading of lower leaves by upper leaves. Shadows can reduce foliage reflectance and give the appearance on film of less than 100% ground cover, even though lower leaves on the plants from adjacent rows are touching and interpenetrating (Gerbermann et al., 1976). Reducing the HAG of the sensing platform to less than 10 meters can amplify the influence of canopy architecture (Fritz, 1967). Also, if the field of view is so small that only a small part of the canopy is measured, then random variation in the canopy will not be sufficiently sampled in a single measurement (Colwell, 1974).

2.3.3 Film

Milton (2002) identified three disadvantages to using FCIR photography. First, normal exposure meters on 35 mm cameras are designed to be insensitive to NIR wavelengths, therefore bracketing photos is required to ensure capturing a correct exposure, often resulting in many wasted photographs. Second, it is more difficult to evaluate the scene being photographed, i.e. light conditions, sun angle, etc., when the false color image is so different from the visible image. This could make choosing the correct exposure from a visual assessment of a slide difficult. Third, FCIR film requires special handling, presenting a higher risk of loss or damage than color film and possibly increasing the time between image collection and image development. The film must be stored in a freezer or refrigerator to preserve the emulsions and thawed at least five hours before anticipated use. This presents challenges to scheduling data collection, particularly if weather conditions change between thawing and photography.

2.3.4 Image Analysis

Photographs can be digitized so that the brightness of each pixel within the photograph is quantitatively indicative of the light emanating from an identifiable area on the landscape (Blackmer and Schepers, 1996). Digital images from scanned slides must be saved in a lossless format such as TIF to preserve as much of the photographic data as possible.

Scharf and Lory (2000) found that color-balancing photographs could not be standardized across photos. Several scientists have used a combined process of digitizing FCIR slides and orthorectifying the images with GPS data in image analysis programs (Flower et al., 2001).

Lukina et al., (1999) used a digital camera and an image processing program to compare spectral irradiance readings (671 and 780 nm) of winter wheat at Feekes GS 4 and 5 to pixel brightness values in order to estimate percent vegetation coverage and biomass.

They found that soil color, soil brightness, and variable light conditions required a significant amount of color balancing within the image processing program and no single procedure could be established that would handle all the images at different locations.

Milton (2002) developed a method of using a commercially available digital camera and filter to create ground-based FCIR images. Blackmer et al. (1996a) digitized color aerial photographic transparencies of corn at growth stage R5, and obtained significant correlations between R and G digital pixel values and grain yield, despite variability in background lighting and digitizing technique. Flowers et al. (2001) used a similar digitizing process of color infrared photographs with the ERDAS image analysis program

(ERDAS Imagine v 8.3, Atlanta, GA). They found that NIR pixel values could be used to predict tiller density in winter wheat at GS 25.

2.4 Remote Sensing of Forages

Forage canopies differ from canopies in annual row crop and forest systems by canopy architecture and agronomic management, which presents challenges to the utility of remote sensing in estimating biomass and N concentration. Many grasses have a smaller LAI at maturity than most row crops, particularly turf-type grasses such as bermudagrass. Additionally, the leaf angle from the stem is often more vertical as compared with row crops, such as soybean or corn, which present a more horizontal leaf surface towards incident radiation. Wooley (1971) found higher visible and lower NIR reflectance when sensing the back of a soybean leaf versus the face typically facing the sun. Grasses tend to have similar spectral properties on either side of the leaf. Consequently, lodging conditions could result in much more variable reflectance values for a soybean canopy versus a pure stand of bermudagrass.

Bare soil in annual row crop production is very rare in forage canopies. Even when forages are harvested, stubble and leaf litter often cover the soil such that the effect of bare soil reflectance is less than with conventional cropping systems, which often require a bare soil reference within the field for calibration of remote sensing data. In addition, canopy cover occurs at a faster rate for forage canopies than row crops, particularly for grasses that spread through stolons and rhizomes (e.g., bermudagrass). Colwell (1974)

examined a grass canopy consisting of a representative mixture of several different species on two different colored soils ("light" and "dark") with similar vegetative covers (~37%), and reported R spectral responses nearly three times higher on the light-colored soil than on the dark colored soil. He concluded that R reflectance of grass canopies had a high negative correlation with percent vegetation cover when viewed over a light-toned soil, but was almost insensitive to changes in percent cover when viewed over a dark-colored soil. Sembiring et al. (1998) predicted biomass, N uptake, P uptake, and N concentration from a bermudagrass canopy using a spectral ratio (695 / 405 nm) obtained with an aboveground sensor (PSD1000 spectrophotometer, Ocean Optics Inc.; HAG = 1.5 m) without the need for a bare soil calibration strip. This suggests that plant canopies that have complete vegetative cover, such as sod forming grasses, can be photographed without a bare soil reference.

Forages used in grazing systems can have a wide range of regrowth rates and canopy covers. Factors such as manure and urine distribution from livestock can create a wide range of nutrient levels in a single field. This translates into variable regrowth rates, pigmentation, and plant type (legume vs. grass, warm season vs. cool season, and the presence of weedy species). Physiological processes associated with regrowth following defoliation through harvesting or grazing were dominant influences on reflectance from tallgrass prairie canopies (Turner et al., 1992). Increased grazing pressures on rangeland pastures in Montana resulted in variable relationships between LAI and NDVI, which could not be credited to one specific factor (Aase et al., 1987). In a study of sheep (*Ovis aries* L.) grazing alfalfa (*Medicago sativa* L.) pastures, Mitchell et al. (1990) concluded

that NDVI was preferable to RVI because it was more sensitive to low levels of phytomass. They concluded that NDVI measurements could be used to identify a threshold phytomass level, below which continued grazing caused a decrease in lamb weight gain. Carnegie et al. (1974) conducted a study in California using a ratio of LANDSAT MSS7/MSS5 bands (800 to 1100 nm / 600 to 700 nm) on an annual grassland canopy and found that the ratio peaked during the greatest periods of forage production. Additionally, they noticed a drop in the ratio during the drying off period following peak growth and concluded that all annual vegetation had died when the ratio did not change.

Variable rates of regrowth often result in a mixture of green and brown (senescing) tissue. Tucker (1979) conducted a study on blue grama grass (*Bouteloua gracilis*) at three different canopy compositions (80% live/ 20% dead, 52% live / 48% dead, and 100% dead) evaluated using 17 different spectral indices obtained through LANDSAT satellite imagery. In the 80% live canopy, G, R, NIR, RVI, DVI, and NDVI were strongly correlated with dry biomass and total chlorophyll ($r^2 > 0.74$). Correlations between NDVI and total biomass ($r^2 = 0.84$) were strong, however when total dry biomass was fractioned into brown and green components, dry green biomass had a much higher correlation ($r^2 = 0.91$) with NDVI than dry brown biomass ($r^2 = 0.56$), indicating that the amount of dead tissue affects the sensitivity of the index. When the canopy was 52% green, correlations for all spectral indices weakened, however when the canopy was 100% dead, correlations strengthened for R and NIR. Dead leaves are the largest contributor to leaf litter and thatch in forage canopies, which can alter reflectance at low canopy covers.

Pastures that are rotationally grazed versus grown for hay exhibit different levels of productivity. A plant that is defoliated on a regular basis is in a constant state of active photosynthesis as opposed to a canopy that is grown for hay, which reaches maturity and puts the energy from radiation into storage versus leaf growth. Using FCIR aerial photography, Curran (1983) found that NDVI measured from a pasture that had been grazed the previous year was higher than an adjacent pasture that had not been grazed even though the standing green biomass on the ungrazed pasture was thirteen times greater. He credited this to increased productivity of the green biomass from the grazed pasture since it had spent more time in active regrowth than the ungrazed pasture. His observations suggest some challenges to interpreting aerial photography from rotational grazing farm systems that would not be encountered when sensing annual crops.

Considerably less research has been done on using remote sensing to evaluate pasture systems as compared to food crops and forests. This study attempts to discriminate how the interpretation of FCIR imagery changes when used to estimate biomass and N concentration in forage canopies.

Table 1. Spectral bands and indexes investigated using false color infrared film.

Spectral Band or Index	Definition	Reference
Green (G)	490 to 550 nm	†
Red (R)	550 to 700 nm	†
Near Infrared (NIR)	700 to 900 nm	†
Normalized Difference Vegetation Index (NDVI)	$(\text{NIR} - \text{R}) / (\text{NIR} + \text{R})$	Rouse et al., 1973
Normalized Green (NG)	$\text{G} / (\text{NIR} + \text{R} + \text{G})$	
Normalized Red (NR)	$\text{R} / (\text{NIR} + \text{R} + \text{G})$	
Normalized NIR (NormNIR)	$\text{NIR} / (\text{NIR} + \text{R} + \text{G})$	Jain, 1989
Green NDVI (GNDVI)	$(\text{NIR} - \text{G}) / (\text{NIR} + \text{G})$	Gitelson, 1996
Ratio Vegetation Index (RVI)	NIR / R	Jordan, 1969
Difference Vegetation Index (DVI)	$\text{NIR} - \text{R}$	Tucker, 1979

† Kodak Ektachrome Infrared EIR film, Eastman Kodak Co., Rochester, NY.

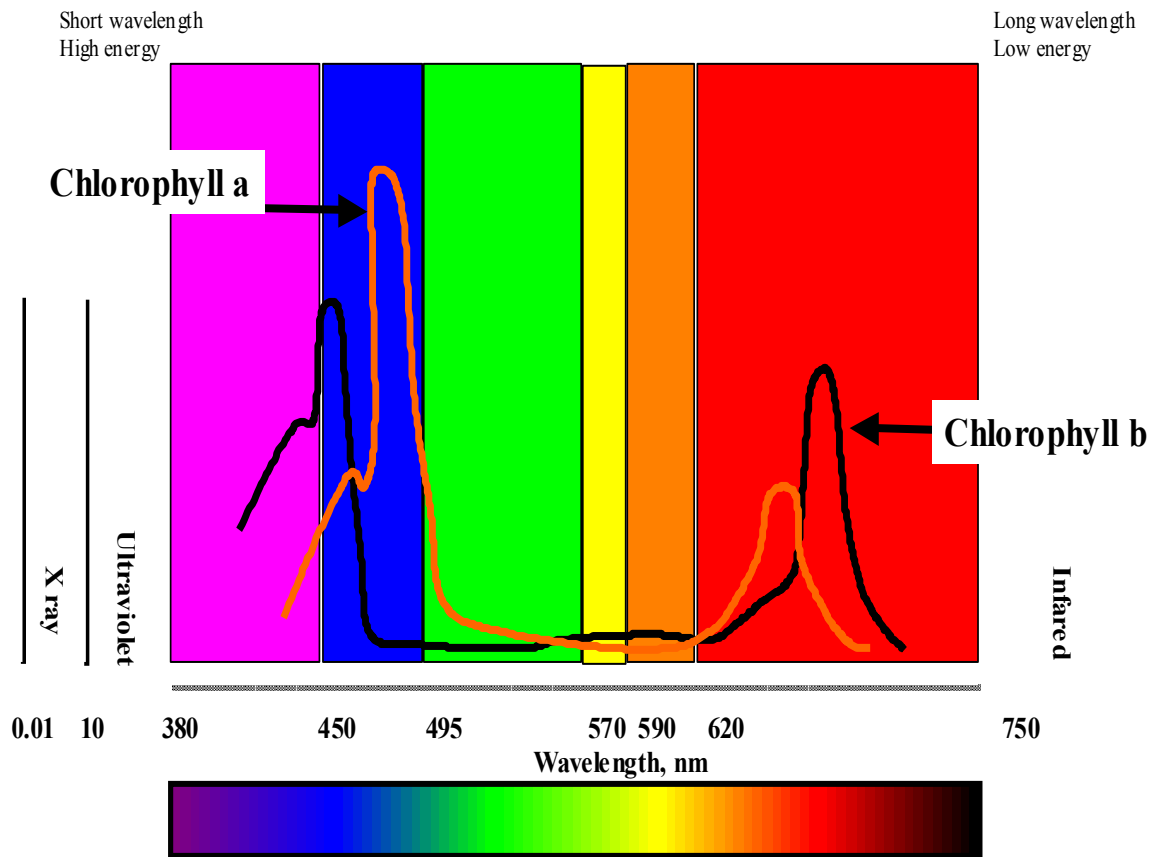


Figure 1. Absorption of light by plant photopigments.

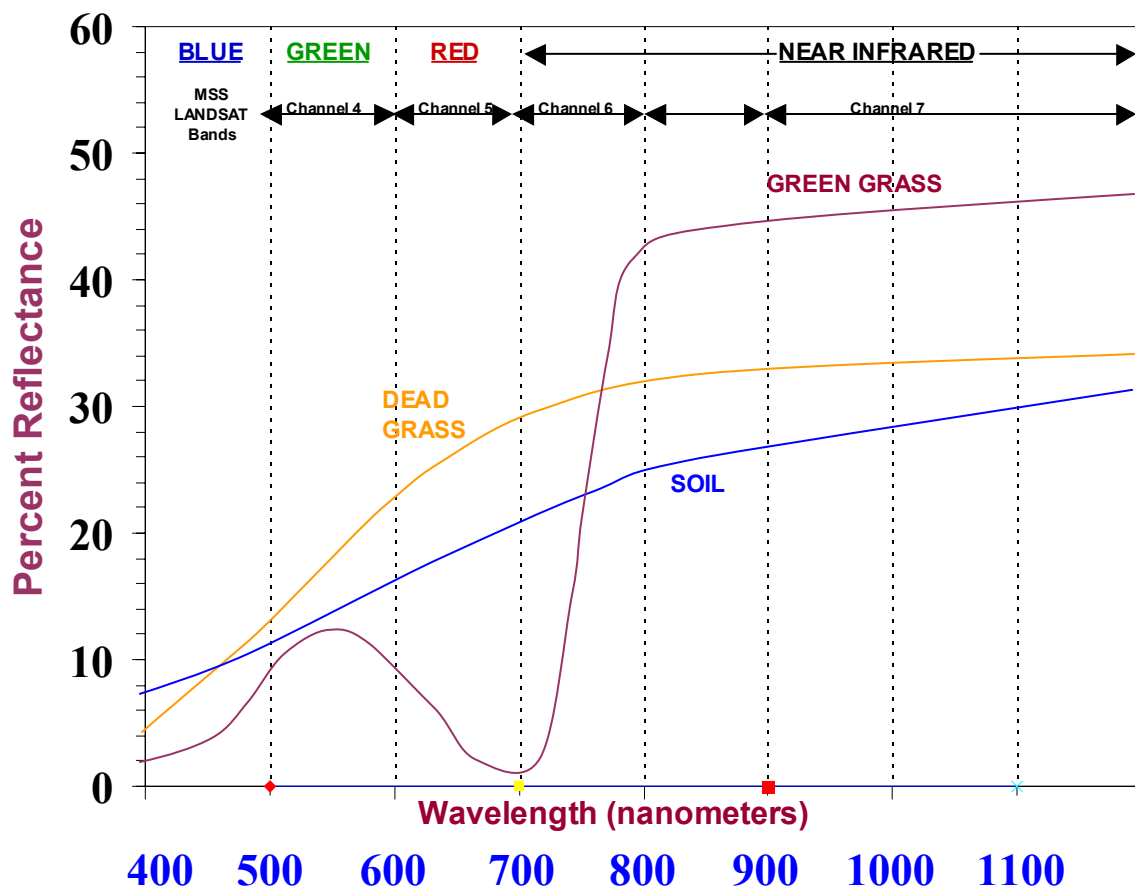


Figure 2. Idealized reflectance patterns of herbaceous vegetation and soil from 0.4 to 1.1 nm (Deering et al., 1975).

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Estimating Forage Biomass and Nitrogen Concentration Using False Color Infrared Aerial Photography

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Abbreviations: FCIR, false color infrared; B, blue; G, green; R, red; NIR, near infrared; BG, bermudagrass; CG, crabgrass; VI, vegetation index; N, nitrogen; VWS, volunteer warm season; NDVI, normalized difference vegetation index; RVI, ratio vegetation index; DVI, difference vegetation index; NormNIR, normalized near infrared; NG, normalized green; NR, normalized red; GNDVI, green normalized difference vegetation index.

Keywords: bermudagrass, crabgrass, false color, infrared, aerial photography, NDVI, normalized green, normalized red, DVI, RVI, effluent, Green NDVI, remote sensing, forage

ABSTRACT

Remotely assessing nitrogen (N) concentration and biomass of grassland canopies can provide land managers and regulatory agencies an opportunity to quickly determine the grassland inventory that is available for grazing animals, wildlife, or soil protection. Limited research has been done examining the utility of false color infrared (FCIR) aerial photography in estimating biomass and N concentration in warm-season forage canopies. The objectives of this study were to determine if a vegetation index (VI) or digital counts in the green (G), red (R), or near infrared (NIR) derived from FCIR aerial photography could be used to estimate biomass and N concentration in bermudagrass (*Cynodon dactylon* L.). The experiment was a three-way factorial treatment design with two N sources (swine effluent and ammonium nitrate [NH₄NO₃]), three forage canopies (bermudagrass [BG], crabgrass [CG] [*Digitaria sanguinalis* L.], and a volunteer warm-season grass canopy composed of 80% crabgrass and 20% forbs [VWS]), and four N rates (0, 224, 449, and 674 kg N ha⁻¹ yr⁻¹). A FCIR aerial photograph was taken in July, 2000 at an altitude of 854 meters. Biomass, N concentration, and N uptake were measured and regressed against spectral variables. Differences due to N source affected the relationship between biomass and GNDVI in BG, and many of the relationships in VWS, but GNDVI was a consistently strong estimator of N uptake for all forage species. Biomass was best estimated by NIR digital counts ($R^2 = 0.82$) in BG, NDVI ($R^2 = 0.54$) in CG, and Normalized NIR ($R^2 = 0.86$) in VWS. N concentration was best estimated by NDVI ($R^2 = 0.62$) in BG, NIR digital counts ($R^2 = 0.56$) in CG, and G digital counts (R^2

= 0.63) in VWS. Results indicate that forage biomass, N concentration, and N uptake can be estimated from FCIR aerial photography, but further research is needed to investigate the effects of N source.

INTRODUCTION

Remotely assessing nitrogen (N) concentration and biomass of grassland canopies can provide land managers and regulatory agencies an opportunity to quickly determine the grassland inventory that is available for grazing animals, wildlife, or soil protection.

Unlike instruments that measure reflectance directly, false color infrared (FCIR) photography captures all the light in a given scene regardless of source (e.g. reflectance, transmittance, and scatter) onto a film emulsion, which can be scanned and digitized so that the brightness of each pixel within the photograph is quantitatively indicative of the light emanating from a given area on the landscape (Blackmer and Schepers, 1996).

Researchers have used FCIR aerial photography to estimate ground cover in row crops, detect winter injury in alfalfa (*Medicago sativa* L.), and predict forage biomass and N content (Gerbermann et al., 1976; Wallen et al., 1977; Ryan et al., 1989). Research has shown that green plant biomass has a positive relationship with near infrared (NIR) reflectance and a negative relationship with red (R) and blue (B) reflectance, wavelengths associated with chlorophyll absorbance (Tucker and Maxwell, 1976). Chlorophyll is positively associated with N concentration (Wolfe et al., 1988). Reflectance measurements at 550 nm have been reported to be indicative of chlorophyll and N concentration (Knipling, 1970). Spectral vegetation indices (VIs) derived from photographic green (G) (490 to 550 nm), R (550 to 700 nm), and NIR (700 to 900 nm) have been found to be better predictors of canopy responses than measurements from

individual wavebands (Wanjura and Hatifeld, 1987). Aerial spectral measurements of row crop canopies can be more influenced by soil background, presence of senesced or weedy vegetation, and canopy architecture (Curran, 1981, Scharf and Lory, 2000).

The objective of this study was to determine if a spectral index, NIR, R, or G digital counts derived from a FCIR aerial photograph could be used to estimate biomass and N concentration in a several warm-season forage canopies fertilized with ammonium nitrate (NH_4NO_3) or swine effluent.

MATERIALS AND METHODS

An experiment established in 1998 at the Caswell Farm in Kinston, NC to investigate yields from warm-season forages fertilized with anaerobic lagoon swine (*Sus scrofa domestica*) effluent or NH_4NO_3 , was chosen for aerial photographic analysis in July, 2000. The experiment was a three-way factorial treatment design (2 N sources by 3 forage species by 4 N rates) implemented in a stripped split-plot arrangement with three replications on a Pocalla soil (loamy, siliceous, thermic Arenic Plinthic Paleudults). At the start of the experiment, the soil had a pH of 4.9, 173 kg P ha⁻¹, and 35 kg K ha⁻¹ (Mehlich, 1984). The main plot factor was N source: swine effluent or NH_4NO_3 . The stripped plot factor was forage species: bermudagrass (BG) (*Cynodon dactylon* L. 'Coastal') overseeded with annual ryegrass (*Lolium multiflorum*), crabgrass (CG) (*Digitaria sanguinalis* L. 'Red River') overseeded with ryegrass, and volunteer warm season (VWS) (80% crabgrass and 20% forbs). Subplots consisted of four N rates (0,

224, 449, and 674 kg N ha⁻¹) split into multiple target applications per year applied in February (15%), March (14%), April (5%), May (7.5%), June (17%), July (24%), September (12.5%), and November (5%) for BG and CG, and February (15%), April (30%), May (11%), June (9%), August (18%), September (11%), and November (6%) for VWS.

Nitrogen rates for the VWS plots deviated from the annual schedule due to field error, and resulted in a significant over-application of NH₄NO₃ in May. Phosphorous (P) was applied to all zero rate and NH₄NO₃ plots at 58 kg P ha⁻¹ in March. Potassium (K) was applied in split applications to all zero rate and NH₄NO₃ plots at 58 kg K ha⁻¹ in March and 112 kg K ha⁻¹ in September. Lime was applied in May to all plots according to soil test recommendations at a rate of 2.2 Mg ha⁻¹. Soil tests indicated the need for sulfur (S) which was applied in May to all plots at 22 kg S ha⁻¹. The nutrient concentration of effluent for the N applications immediately prior to the July harvest was: 278 mg N kg⁻¹, 96 mg P kg⁻¹, and 221 mg K kg⁻¹. Based on the annual fertilization schedule, the amount of N applied since the previous harvest was 38 kg ha⁻¹ of NH₄NO₃ and 36 kg ha⁻¹ of N in the form of effluent on the 224 kg ha⁻¹ (base) rate plots. The average rainfall since the previous harvest (42 days) was 91 mm.

Forage biomass was harvested from an area of 5.2 m² leaving a stubble height of 5 cm using a Haldrup (Wintersteiger, Salt Lake City, Utah) sicklebar forage harvester.

Subsamples of each harvest were weighed, dried in a forced-air oven at 65 °C for 24 to 48 hours, reweighed to determine tissue moisture, ground to 1-mm, thoroughly mixed,

and submitted for N analysis. Samples were analyzed for N concentration using a Perkin Elmer 2400 CHN Elemental Analyzer (Perkin Elmer Corp., Norwalk, CT.). Biomass was calculated as the dry matter yield above cutting height. Nitrogen uptake was determined by multiplying N concentration and biomass.

Calibration markers were placed on corners of the experimental area to locate the plots on the digital image. Latitude and longitude for the calibration markers were determined using a differential global positioning system (DGPS) receiver (Trimble AG 132, Trimble Navigation, Ltd, Sunnyvale, CA.). One day prior to harvest, the experimental area was photographed at an altitude of approximately 854 m with a 35-mm manual SLR camera (Nikon 6000, Nikon, Inc., Melville, NY) fitted with a Kodak WRATTEN Gelatin Filter No.12 (yellow) to absorb blue radiation. Kodak Ektachrome Color Infrared EIR film (Eastman Kodak Co., Rochester, NY) was used for all photography. The film was processed into a false color positive slide, which was digitized to a tagged image file format (TIFF) using a slide scanner (Konica Q-Scan, Konica Corp., Mahwah, NJ.) with a resolution of 47 pixels per mm (1000 dpi). The result was a 24-bit RGB image (8 bit red, 8 bit green, 8 bit blue) with the brightness of each primary color value represented by a RGB digital count within the range of 0 to 255, whereby NIR was represented by red, R was represented by green, and G was represented by blue. The photograph was orthorectified using ERDAS Imagine (ERDAS, Atlanta, GA) (Flowers et al., 2001). The harvest area of each plot, representing a ground area of 5 m², was selected for spectral analysis.

Pixel brightness values for the three wavebands represented in the film (R, G, and NIR) were determined using ERDAS Imagine Version 8.3.1 (ERDAS, 1997) from the digitized photographed harvest area. Foliar injury was observed in the field and on the photograph within the high N rate CG and VWS plots that received NH_4NO_3 . These plots were removed from the data analysis to avoid sources of variation that were unrelated to the original hypotheses. As a result, the high N rate treatment receiving NH_4NO_3 was eliminated for CG and VWS.

In addition to the digital counts in each band, a number of ratios were derived and used for statistical analysis with the crop response variables (Table 1). Analysis of variance was performed using SAS Version 8 (SAS, Cary, NC) on the spectral and crop response variables (biomass, N concentration, and N uptake) to determine interactions between N rate, N source, and forage species. A sequence of linear versus quadratic models were determined using Proc GLM. Proc REG was used to determine coefficients of determination when a linear model was adequate. Analysis of variance in Proc GLM was used to determine the effect of N source on the relationship between a crop response variable and a spectral variable. Crop response variables are presented graphically as the dependent variable in order to test the ability of the raw digital count or spectral index to estimate canopy characteristics. For the purposes of this paper, correlations are defined as strong ($R^2 > 0.7$), moderate ($0.5 < R^2 < 0.7$), and weak ($R^2 < 0.5$).

RESULTS AND DISCUSSION

Agronomic response to N rate and N source

Differences among forage species prohibited a combined analysis. Excess application of NH_4NO_3 on VWS canopies precluded its inclusion in an analysis of agronomic responses to N rate and N source. There was a N source by N rate interaction for BG biomass, N concentration, and N uptake, and for CG N uptake (Figure 3). Fertilization with NH_4NO_3 generally resulted in higher mean values for the crop response variables. Differences due to N source may have been a result of fertilizer composition. While additional P and K were applied to NH_4NO_3 plots, the application rates were based on soil tests from the 224 and 448 kg ha^{-1} rate plots, and the amount of P and K applied with each effluent application varied considerably because of varying effluent composition. Additionally, NH_4NO_3 was applied in a prilled form and may not have been plant available until moisture was present, while effluent was an aqueous solution, much of which was immediately plant available.

Biomass in BG canopies increased in response to increasing N and appeared to reach a plateau with both N sources near 5 Mg ha^{-1} (Figure 3). Conversely, BG N concentration remained relatively constant with increasing N, until biomass had plateaued, whereby N concentration increased more steeply. Biomass was still increasing in CG plots at the highest N rate while N concentration decreased across N rates. Nitrogen uptake increased with increasing N for both species, with a somewhat greater increase occurring for CG with NH_4NO_3 .

Spectral response to N rate and N source

Green and NIR digital counts, and NormNIR, NDVI, RVI, DVI, GNDVI, and NG VIs from BG canopies all increased and plateaued at the higher N rates, while R and NR decreased with N rate (Appendix 6.2). In general, spectral trends across N rates for CG canopies were similar to those observed in BG, however there was a greater difference observed between the two N sources. Source of N resulted in two statistically different ($p < 0.05$) trends across N rates for G and R digital counts, and NormNIR, DVI, RVI, NR, NG, and GNDVI VIs, with greater values associated with plots fertilized with NH_4NO_3 than effluent. Positive trends were observed for NormNIR, DVI, RVI, NDVI, and GNDVI, while negative trends were observed for R, NG, and NR. Digital counts for NIR did not appear to vary across N rates.

Relationships between spectral and crop response variables within forage species

Bermudagrass. Coefficients of determination (R^2) for the relationships between nonnormalized (raw) digital counts and crop response variables were generally moderate to strong for NIR and R and insignificant for G (Table 2). Relationships between NIR and biomass are consistent with those reported in other studies using FCIR aerial photography (Flowers et al., 2002). With biomass ranging from 1.1 to 5.0 Mg ha^{-1} , senescent tissue may have contributed to weak correlations with G and R digital counts which respond primarily to plant pigments. Greater R digital counts were associated with lower N concentrations and biomass in the canopy. Increased R reflectance has been

associated with lower levels of chlorophyll and N in the plant tissue (Colwell, 1974 [oats]; Hinzman et al., 1986 [wheat]; Blackmer et al., 1994 [corn]).

Dividing NIR digital counts by the sum of the bands (NormNIR) did not improve the correlation with biomass, however it strengthened the relationships with N concentration and N uptake dramatically (Figure 4, Table 2). Flowers et al. (submitted) found similarly strong relationships between NormNIR and tiller density for wheat canopies at GS 25 and N concentration at GS 30 sensed with FCIR aerial photography. Figure 5 showed similar improvements as NormNIR versus NIR, but was slightly better at estimating biomass and N concentration than NormNIR. The vegetation indices were generally stronger estimators of the crop response variables than the raw digital counts.

Crabgrass. Coefficients of determination (R^2) for some of the relationships between raw digital counts and crops response variables were slightly stronger than those found in BG (Table 2). Negative weak correlations were found between G digital counts and biomass and N concentration, however, there was moderate negative correlation between G and N uptake. Red digital counts were better correlated to biomass and N uptake than relationships observed in BG.

Digital counts for NIR were uncorrelated with CG biomass and N uptake and negatively correlated with N concentration, indicating that the greatest amount of N in the plant tissue was associated with lower digital counts. These results correspond with numerous observations by other researchers that NIR reflectance is negatively associated with N

concentration and positively associated with biomass (Walburg et al., 1982; Colwell, 1974). The range in CG biomass (0.34 to 5.55 Mg ha⁻¹) was less than that observed for BG, indicating that NIR digital counts may not be good estimators of biomass under 5 Mg ha⁻¹. The elimination of the high N rate plots fertilized with NH₄NO₃ may have prevented the relationship from looking similar to that observed with BG. Normalized NIR and NDVI were considerably better than NIR digital counts at estimating biomass and N uptake, but weaker for estimating N concentration (Figures 5).

Volunteer Warm Season. Green and R digital counts had moderate to strong negative correlations with VWS biomass and N uptake, indicating that the visual brightness of the canopy decreased as biomass and N concentration increased (Table 2). The NIR digital counts had no correlation with any of the crop response variables. While the VWS canopy was observed to be 80% crabgrass, the difference in cultivar (volunteer vs. Red River) and the presence of some warm-season forbs (e.g. yellow foxtail and goosegrass) in the VWS canopies may have contributed to the higher correlations found in the VWS canopies. Figure 5 illustrates similar linear relationships between GNDVI and N uptake for VWS and CG canopies, with VWS having a slightly stronger correlation.

Vegetation indices tended to have stronger correlations than raw digital counts for estimating biomass, whereby NormNIR (Figure 4) was the best estimator. Relationships between biomass and G, NormNIR, RVI, and NG were quadratic, reaching a plateau around 4 Mg ha⁻¹. Nitrogen uptake was generally best predicted by GNDVI, and it was

generally better predicted than either biomass or N concentration for all forage species (Figure 6, Table 2).

Effects of N source on Relationships between Spectral and Crop Response Variables

Source of N had an effect on the relationships between biomass and GNDVI for BG and VWS (Figure 7). There was a wider range of biomass for BG plots fertilized with NH_4NO_3 , and a moderate positive linear correlation, while plots fertilized with effluent had an average of 0.81 Mg ha^{-1} less mean biomass than the equivalent treatments fertilized with NH_4NO_3 . The opposite trend was noticed in VWS canopies where biomass seemed to plateau at 4 Mg ha^{-1} for plots fertilized with NH_4NO_3 , which could be due to the removal of high N rate plots fertilized with NH_4NO_3 . Additionally, effects of N source were found in VWS canopies for the relationships of N concentration with NormNIR, NG, and GNDVI, and of N uptake with NDVI, RVI, DVI, and NR (Table 2). While differences due to N source were noticed in BG ($n = 12$ for both N sources), results found in VWS may be due in part to a loss of almost half of the plots fertilized with NH_4NO_3 as a result of foliar injury ($n = 7$ for NH_4NO_3 , $n = 12$ for effluent). Since plots fertilized with NH_4NO_3 produced more biomass for the other forage species, the loss of these points could have affected indices that are sensitive at high levels of vegetation. Even with a reduced number of data points, relationships with NH_4NO_3 were not always better than those with effluent. Coefficients of determination were greater for plots fertilized with effluent for half of the relationships affected by N source in VWS canopies

and the only relationship affected by N source in BG canopies, indicating that effects due to N source did not affect the relationships between spectral and crop response variables consistently (Figure 7). Effects of different N sources within the same photograph have not been documented to the author's knowledge. These results suggest that different relationships may be needed to estimate crop response variables using remote sensing for canopies that have been fertilized with different N sources, such as swine effluent versus NH_4NO_3 .

CONCLUSIONS

Significant relationships between spectral and crop response variables were found, but these differed among forage species. Considering raw digital counts, biomass was best estimated by NIR in BG, by R in CG, and by G in VWS canopies. Nitrogen concentration was best estimated by R digital counts in BG, NIR digital counts in CG, and G digital counts in VWS. Nitrogen uptake was best estimated by NIR digital counts in BG and G digital counts in CG and VWS.

Normalization of the raw digital counts generally improved correlations for all three forage species except in the relationship between biomass and NIR digital counts in BG. Biomass was similarly predicted by NormNIR, NDVI, and DVI for BG, CG, and VWS. Nitrogen concentration was similarly predicted by NDVI and DVI across forage species, while N uptake was similarly predicted by NDVI, NormNIR, DVI, and GNDVI. Nitrogen concentration was best estimated in BG, less strongly so in CG, and poorly if at

all in VWS. Spectral response to N concentration is difficult to isolate due to the confounding factor of biomass, however the strong correlations between spectral data and N uptake appear to provide reliable information on the N status of the canopy.

Nitrogen source affected the relationship between biomass and GNDVI in BG, and many of the relationships in VWS. These results indicate that additional research is needed to characterize the potential effects of N source on the relationships between spectral and crop response variables. Green NDVI was a consistently strong estimator of N uptake for all forage species, irrespective of N source. The robustness of GNDVI to a mixture of species and N sources within a single aerial photograph may be due to the fact that it omits the R digital information. Blackmer et al. (1996) found that R digital counts obtained from one aerial photograph of four corn hybrids (R5 growth stage) were significantly affected by variety.

These results indicate that forage biomass, N concentration, and N uptake can be estimated from FCIR aerial photography. Green NDVI was strongly correlated with biomass and N uptake for all forages, and was insensitive to N source. Furthermore, GNDVI was a sensitive predictor of biomass and N uptake in a pure stand (BG) and a forage mixture (VWS). These two conditions represent typical forage composition in nutrient application sites and livestock pastures. Limited extrapolation can be made on this data due to a lack of site and harvest replication, however the results are promising enough to warrant further study. Using FCIR aerial photography as a point-in-time assessment of crop response variables such as biomass, N concentration, and N uptake

can be strengthened by further research to quantify these relationships among cool season forage mixtures, rotational grazing systems, and various stages of growth.

ACKNOWLEDGEMENTS

We thank TJ Holliday and Pete Thompson for their field support. This research was funded in part by North Carolina Department of Environment and Natural Resources.

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Table 2. Coefficients of determination (R^2) for relationships between spectral and crop response variables for bermudagrass, crabgrass, and volunteer warm season canopies harvested in July, 2000 in Kinston, NC.

	Spectral Variable									
	G	R	NIR	NDVI	NormNIR	RVI	DVI	NR	NG	GNDVI
<u>Bermudagrass</u>	R^2									
Biomass	NS	-0.35	0.82	0.77	0.74	0.75	0.81	-0.77	NS	§
N Concentration	NS	-0.52	0.14	0.62 [†]	0.55	0.62 [†]	0.63 [†]	-0.60 [†]	NS	0.61
N Uptake	NS	-0.51	0.59	0.79	0.81	0.80	0.80	-0.76	NS	0.79
<u>Crabgrass</u>										
Biomass	-0.25	-0.47 [†]	NS	0.54 [†]	0.55 [†]	0.54 [†]	0.40	-0.54 [†]	-0.34	0.59 [†]
N Concentration	-0.24	NS	-0.56	0.44 [†]	0.34 [†]	0.44 [†]	0.44 [†]	-0.45 [†]	NS	NS
N Uptake	-0.59	-0.53	NS	0.57	0.68	0.56	0.58	-0.46	-0.64	0.76
<u>Volunteer Warm Season</u>										
Biomass	-0.77 [†]	-0.69	NS	0.75	0.86 [†]	0.82 [†]	0.78	-0.69	-0.74 [†]	§
N Concentration	-0.63 [†]	-0.25	NS	0.20	§	0.22	NS	NS	§	§
N Uptake	-0.84	-0.77	NS	§	0.88	§	§	§	-0.76	0.92

[†] indicates a quadratic relationship

NS, not significant

- indicates a negative correlation

§ denotes significant N source interaction

Table 3. Coefficients of determination (R^2) between vegetation indices and crop response variables for volunteer warm season canopies fertilized by (1) swine effluent (n = 12) and (2) NH_4NO_3 (n = 7) at Kinston, NC in July, 2000.

Crop response variable	Vegetation Index													
	NDVI		NormNIR		RVI		DVI		NR		NG		GNDVI	
	N Source													
	1	2	1	2	1	2	1	2	1	2	1	2	1	2
-----R ² -----														
Biomass													0.84	0.96 [†]
N Concentration			‡	‡							‡	-0.59	‡	0.84 [†]
N Uptake	0.87	0.82			0.86	0.80	0.88	0.83	-0.81	-0.74				

- indicates a negative correlation

[†] consists of only 2 replications

‡ eliminated due to tissue injury

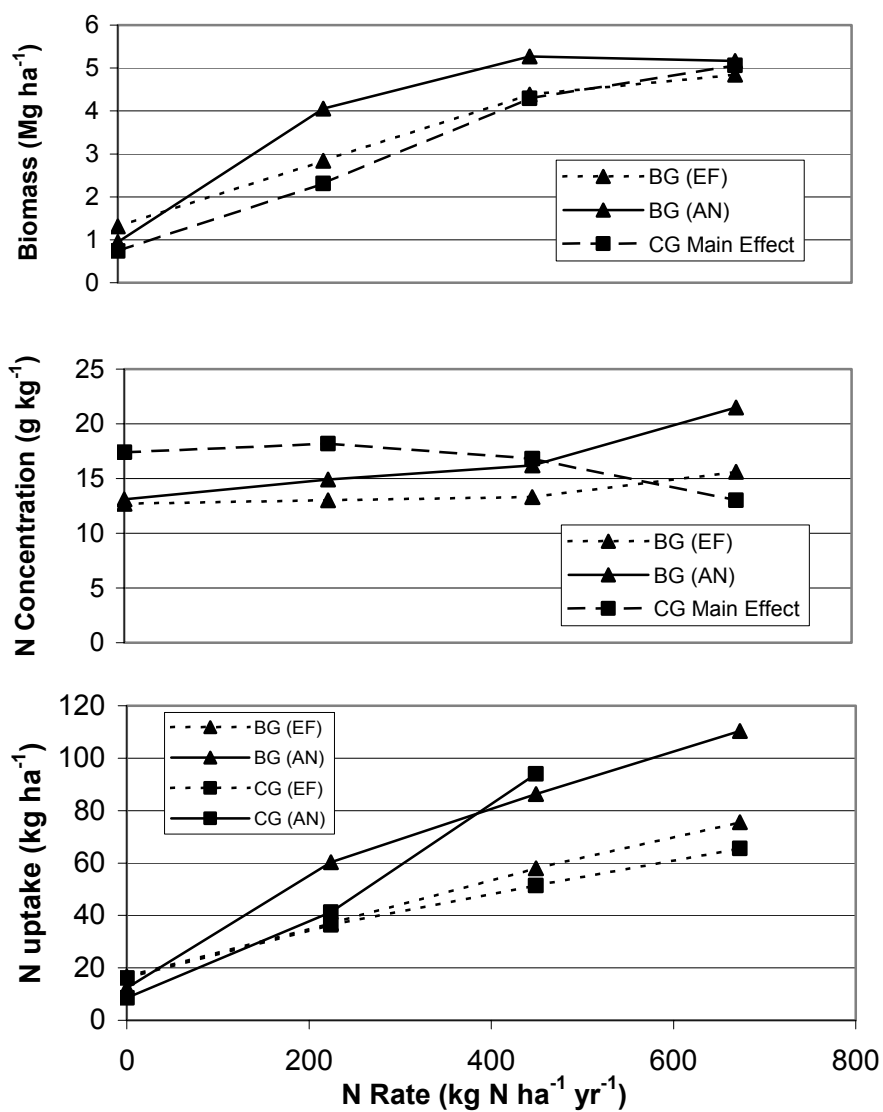


Figure 3. Effects of N on biomass, N concentration, and N uptake for bermudagrass (BG) and crabgrass (CG) fertilized with swine effluent (EF) and NH₄NO₃ (AN) at Kinston in July, 2000.

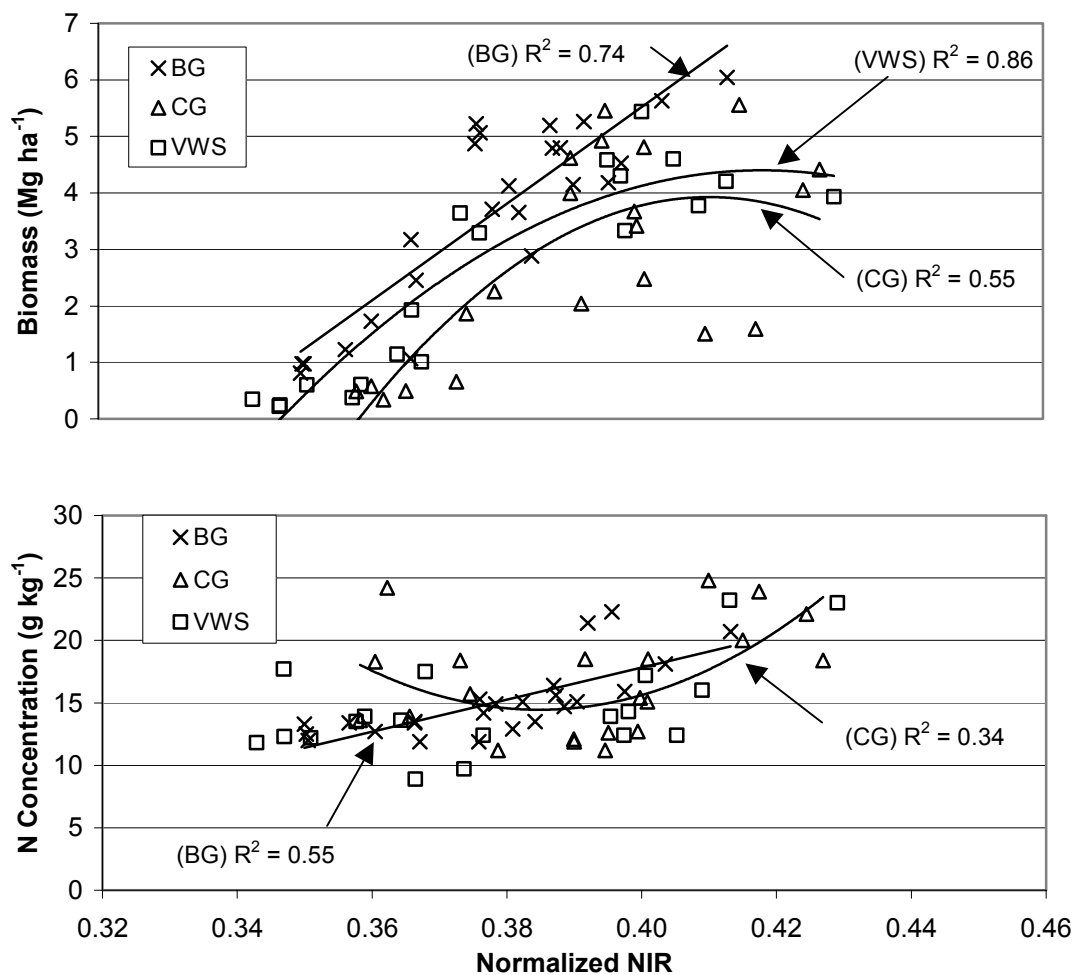


Figure 4. Relationship of Normalized NIR (NormNIR) with biomass and N concentration for bermudagrass (BG), crabgrass (CG), and volunteer warm season (VWS) canopies harvested in July, 2000 at Kinston.

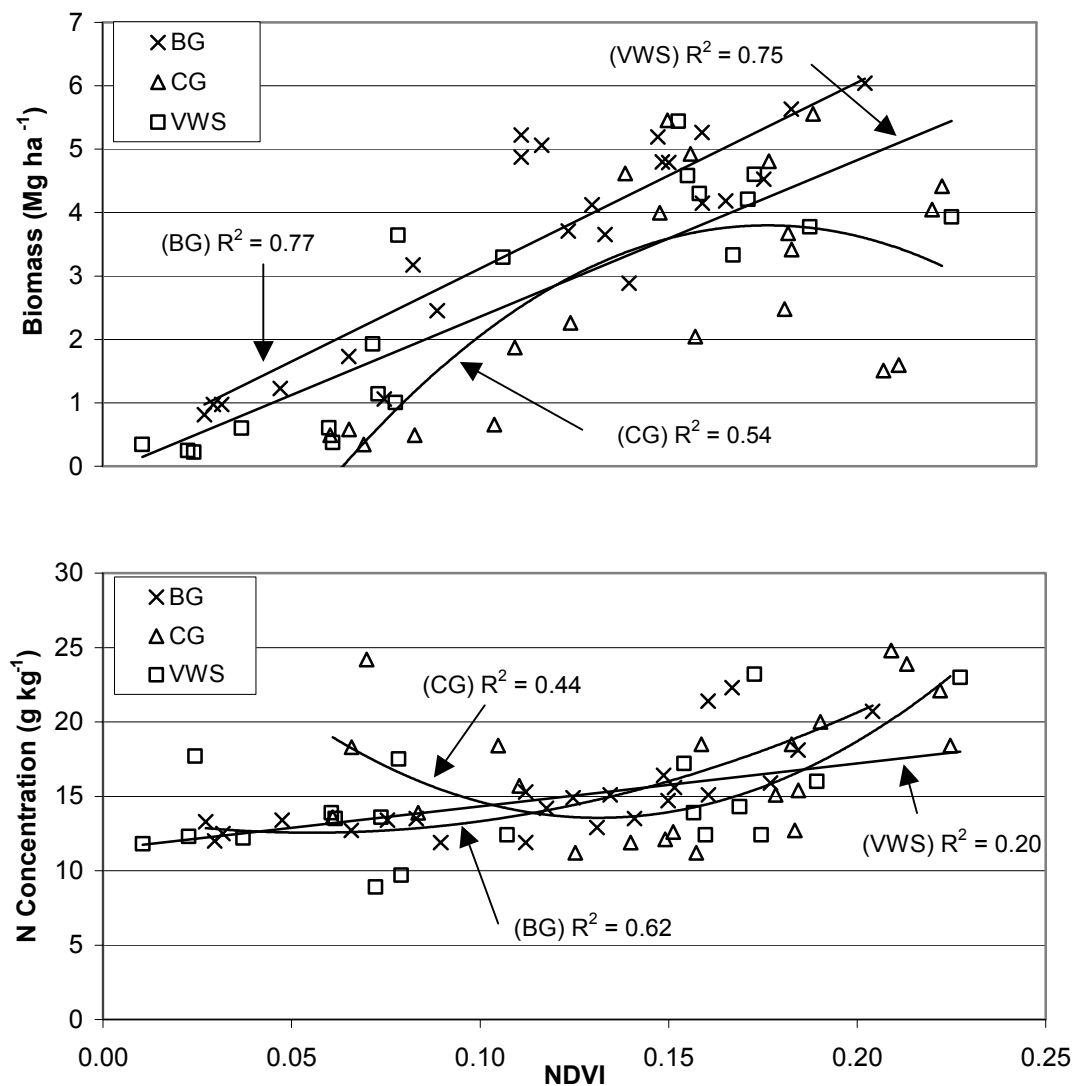


Figure 5. Relationships of NDVI with biomass and N concentration of bermudagrass (BG), crabgrass (CG), and volunteer warm season (VWS) canopies harvested in July, 2000 at Kinston, NC.

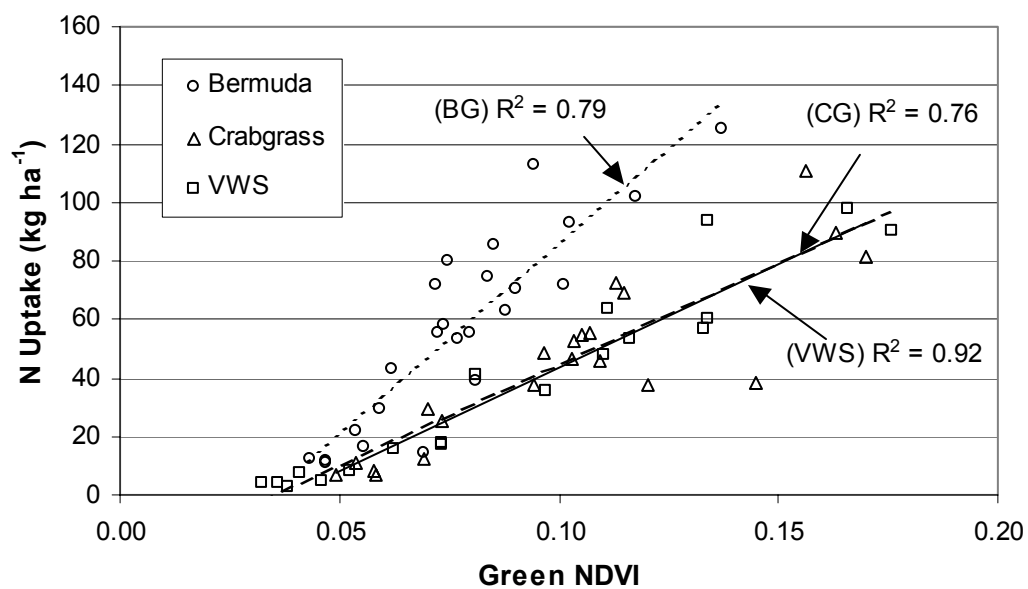


Figure 6. Relationships of Green NDVI with N uptake of bermudagrass (BG), crabgrass (CG), and volunteer warm season (VWS) canopies harvested in July, 2000 at Kinston, NC.

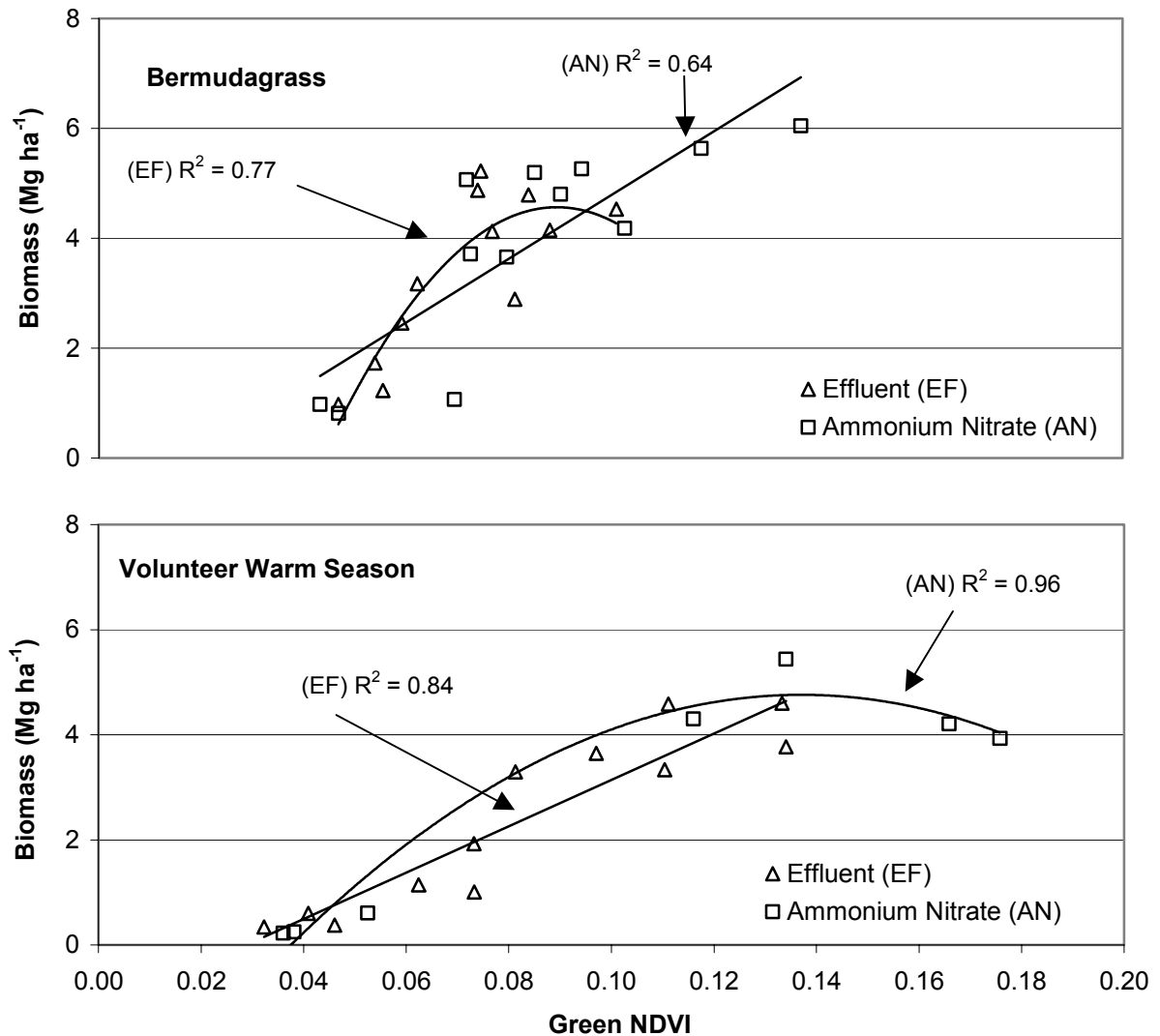


Figure 7. Effects of N source on the relationships between biomass and Green NDVI for bermudagrass (BG) and volunteer warm season (VWS) canopies harvested in July, 2000 at Kinston, NC.

**Estimating Nitrogen Concentration in Bermudagrass (*Cynodon dactylon* L.) at
Similar Biomass Using Ground-based False Color Infrared Photography**

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Abbreviations: FCIR, false color infrared; B, blue; G, green; R, red; NIR, near infrared; BG, bermudagrass; NDVI, normalized difference vegetation index; RVI, ratio vegetation index; DVI, difference vegetation index; NormNIR, normalized near infrared; NG, normalized green; NR, normalized red; GNDVI, green normalized difference vegetation index; VI, vegetation index; NH_4NO_3 , ammonium nitrate; N, nitrogen.

Keywords: bermudagrass (*Cynodon dactylon* L.), biomass, nitrogen, remote sensing, photography, false color, infrared

ABSTRACT

Remote assessment of nitrogen (N) concentration and biomass of grassland canopies can provide land managers and regulatory agencies an opportunity to quickly determine the grassland inventory that is available for grazing animals, wildlife, or soil protection. Limited research has been done examining the utility of ground-based false color infrared (FCIR) photography in estimating N concentration in mature forage canopies. The objectives of this study were (i) to determine if a vegetation index (VI) or digital counts in the green (G), red (R), or near infrared (NIR) could be used to estimate N concentration in a mature canopy of bermudagrass (*Cynodon dactylon* L.) at similar biomass and (ii) to determine the effects of different moisture levels in the plant tissue at similar biomass levels on the utility of these spectral variables. The experiment was a two-way factorial treatment design with three irrigation treatments (no irrigation, irrigated for 25 minutes 24 hours before harvest, and irrigated for 90 minutes 24 hours before harvest) and five N rates (0, 11, 22, 45, and 90 kg N ha⁻¹) applied eleven days before harvest and implemented in a split-plot arrangement with three replications. Data were collected in September, 2000 in Raleigh, North Carolina on a well-established bermudagrass sod at early heading. Photographs were taken with a 35-mm camera elevated 1.83 meters above the ground and spanned the area harvested (0.25 m²). Dry biomass, N concentration, N uptake, tissue color, and soil moisture were measured for each plot. Differences in N concentration at similar biomass were successfully established, but different tissue moisture contents were not. Significant, but weak correlations ($R^2 < 0.28$) were found between N concentration and NIR, NormNIR, DVI,

NG, and GNDVI. When replications were analyzed as sites and irrigation blocks as replications within sites, there was a site \times VI interaction between N concentration and NG and GNDVI that resulted in stronger relationships ($R^2 = 0.38$ to 0.57) for two of the three sites. Strong relationships between N uptake and DVI ($R^2 = 0.83$) and NormNIR ($R^2 = 0.73$) were found at Site 1. Changing light conditions could have caused variation among sites, however, the strength of the relationships found in Site 1 indicate that ground-based FCIR photography can be used to estimate N concentration and N uptake.

INTRODUCTION

Remotely assessing nitrogen (N) concentration and biomass of grassland canopies can provide land managers and regulatory agencies an opportunity to quickly determine the grassland inventory that is available for grazing animals, wildlife, or soil protection. Unlike instruments that measure reflectance directly, false color infrared (FCIR) photography captures all the light in a given scene regardless of source (e.g. reflectance, transmittance, and scatter) onto a film emulsion, which can be scanned and digitized so that the brightness of each pixel within the photograph is quantitatively indicative of the light emanating from a given area on the landscape (Blackmer and Schepers, 1996). Researchers have used FCIR aerial photography to estimate ground cover in row crops, detect winter injury in alfalfa (*Medicago sativa* L.), and predict forage biomass and N content (Gerbermann et al., 1976; Wallen et al., 1977; Ryan et al., 1989). Ground-based FCIR photography has the potential to minimize variability associated with atmospheric conditions that results from aerial or satellite platforms (Scharf and Lory, 2000).

Moisture stress causes changes in leaf water content and structure, which subsequently causes differences in the spectral signature of a canopy, particularly spectral reflectance in the G and IR bands (Gausman, 1974, Myers, 1975). Our first objective was to create a situation where N concentration differences were present without a significant variation in biomass and determine if nonnormalized digital counts or vegetation indices (VIs) derived from ground-based FCIR photography could be used to detect N concentration. Our second objective was to determine the effect of leaf water content on interpreting such ground-based FCIR photographs.

MATERIALS AND METHODS

The experimental area consisted of a well-established bermudagrass sod, located at the North Carolina Agricultural Research Service Unit, Raleigh, NC, on an Appling soil (fine, kaolinitic, thermic Typic Kanhapludults) with a pH of 6.1, 286 kg P ha⁻¹, and 89 kg K ha⁻¹ (Mehlich, 1984). The experiment was a two-way factorial treatment design (3 irrigation treatments x 5 N rates) implemented in a split-plot arrangement with three replications. The main plot factor was irrigation (0, 25, and 90 minutes) applied 24 hours before harvest. The split-plot factor consisted of five levels of ammonium nitrate (NH₄NO₃) at 0, 11, 22, 45, and 90 kg N ha⁻¹ applied eleven days before plots were photographed and harvested. Main plot dimensions were 14 by 8 m; split plot dimensions were 14 by 1.83 m; split-split plot dimensions were 1.83 by 1.83 m.

The experimental area was cut to a height of 5 cm on August 1, 2000 (six weeks before photography) and allowed to accumulate biomass until the forage canopy reached the boot to early heading growth stage in September. Nitrogen treatments were imposed eleven days prior to harvest to allow enough time for N assimilation to result in a chlorophyll response, without resulting in a significant change in biomass among N rates. Plots were irrigated with 19 mm diameter soaker hoses staked down within each plot such that the entire harvest area (0.25 m^2) received consistent moisture for the application time. Natural rainfall (89 mm) between N application and harvest occurred within the first five days post N application. Visual color scores were assigned to each plot at harvest with 1 representing light, yellowing leaf tissue and 10 representing dark green leaf tissue. Botanical separations and amount of inflorescence in each plot were determined post harvest.

Eleven days after N application and just before harvest (September 12, 2000), plots were photographed with a 35-mm SLR manual camera (Model OM-1, Olympus America Inc., Melville, NY) elevated 1.83 m above the ground and leveled for a nadir view over a ground area of 0.25 m^2 . Kodak Ektachrome Color Infrared EIR film (Eastman Kodak Co., Rochester, NY), which responds to Blue, G, R, and NIR wavelengths, was combined with a yellow filter (Kodak Wratten Gelatin Filter No.12) to absorb blue radiation. Replication 1 was photographed between 1300 to 1500 h, and replication 2 was photographed between 1500 to 1700 h. Replication 3 was photographed 48 hours later between 1300 to 1500 h.

Forage biomass was harvested from a 0.25 m² quadrat within each plot and immediately weighed. Three soil cores (to 10 cm depth) were taken within the harvest area, mixed, immediately weighed, dried for 12 hours at 105 °C, and reweighed. Forage samples were dried in a forced-air oven at 65 °C for 24 to 48 hours, weighed to determine dry matter, ground to 1 mm, thoroughly mixed, and analyzed for N concentration using a Perkin Elmer 2400 CHN Elemental Analyzer (Perkin Elmer Corp., Norwalk, CT.). Biomass was calculated as the dry matter yield above 5 cm. Nitrogen uptake was determined by multiplying N concentration by biomass.

The film was processed onto a false color positive slide, which was digitized (TIFF files) using a slide scanner (Konica Q-Scan, Konica Corp., Mahwah, NJ) with a resolution of 47 pixels per mm (1000 dpi). The resulting image consisted of a 24-bit RGB image (8 bit red, 8 bit green, 8 bit blue) with each primary color value representing a RGB digital count within the range of 0 to 255. Pixel brightness values for the three color bands represented in the film (R, G, and NIR) were extracted from the digitized files using ERDAS Imagine Version 8.3.1 (ERDAS, 1997). In addition to the digital values in each band, a number of ratios were derived (Table 1) and used to examine relationships with crop response variables (biomass, N concentration, and N uptake).

The data were analyzed using analysis of variance and simple regression in SAS Version 8 (SAS, Cary, NC). Proc GLM was used to identify any irrigation × N rate interactions. Differences referred to hereafter imply significance at $p \leq 0.05$. Regression and

correlation analyses were performed to assess the relationship between spectral and crop response variables. Analysis of variance was used to determine which of the experimental treatments accounted for variability in spectral responses. A sequence of linear versus quadratic models was determined using Proc GLM. Proc REG was used to determine Pearson correlation coefficients when a linear model was adequate. PROC MIXED was used to test for differences among regression lines. Crop response variables are presented graphically as the dependent variables in order to test the spectral indices as estimators of those variables. For the purposes of this paper, correlations are defined as strong ($R^2 > 0.7$), moderate ($0.5 < R^2 < 0.7$), and weak ($R^2 < 0.5$).

RESULTS AND DISCUSSION

Agronomic and spectral response to N rate and irrigation

Nitrogen affected N concentration, N uptake, tissue moisture, and leaf tissue color, but not biomass (Table 4). Irrigation affected soil moisture and amount of inflorescence, but did not affect tissue moisture nor any of the other crop response variables. Mean biomass ranged between 4.09 to 4.56 Mg ha⁻¹ for all N rates, while mean N concentration and N uptake increased with increasing N rates (Figure 8).

Among raw (nonnormalized) digital counts and VIs, differences due to N were found for NIR, NDVI, NormNIR, RVI, DVI, NG, and GNDVI (Table 5), exhibiting a negative trend for NG across N rates and a general positive trend for non-normalized R, G, and

NIR digital counts (Figure 9) and the other VIs across N rates. Irrigation did not affect any of the spectral variables (Table 5).

Relationships between N concentration and spectral indices

Nitrogen concentration and N uptake were weakly correlated with NIR digital counts, NormNIR, DVI, NG, and GNDVI (Table 6). Nitrogen uptake was also weakly correlated with NDVI and RVI. Biomass was weakly correlated with DVI ($R^2 = 0.14$). The range in N concentration (13 to 23 g kg⁻¹) appeared to be sufficient to be detected spectrally. Previous research by the author found moderate correlations ($R^2 > 0.50$) between N concentration and NIR digital counts and several VIs for N concentrations ranging from 12 to 22 g kg⁻¹ from bermudagrass canopies using FCIR aerial photography (Morgan, 2002). Given the late stage of growth of the canopy and the physiology of bermudagrass in September (cooler night temperatures and slower growth rates), it is possible that the N contained in the plant tissue was primarily in forms other than plant pigments. This could explain the lack of correlation between N concentration and R and G raw digital counts, which primarily respond to plant pigments (Gates et al., 1965). Previous studies have shown that when N concentration increases in plant tissue, absorbance in the R and blue wavelengths increases and reflectance decreases (Walburg et al., 1982; Blackmer et al., 1996). When biomass and N concentration are both increasing due to N rates, R digital counts have been observed to be moderately correlated ($R^2 = 0.52$) with N concentration (Morgan, 2002).

The ground-based method of photography was another potential source of variability. Minimizing the proximity of the camera to the canopy such that individual leaves can be seen can amplify effects of leaf orientation and pigmentation (Fritz, 1967).

Photographing plots individually greatly increases the time necessary to photograph the entire experimental area compared to aerial photography, which can capture the entire scene in one photograph. Hinzman et al. (1986) attributed variations in reflectance observed over a two hour time period to changes in solar angle. Changes in solar angle and varying light conditions could have affected our results since it took on average two hours to photograph each replication.

Given the lack of response to irrigation and the temporal separation between replications, the experiment was reanalyzed as a randomized complete block design, whereby each original replication became a 'site' and the main plots became replications within these sites. There was a site x N rate interaction ($p < 0.05$) for the relationship of N concentration with NG and GNDVI. Furthermore, there was a site x VI ($p < 0.05$) interaction for the relationships of N concentration with NG and GNDVI. Coefficients of determination were generated for the relationships between spectral and crop response variables for each 'site' (Table 7). Correlations were stronger for sites 1 and 2, but insignificant for site 3, indicating that this 'site' may have contributed to the weak correlations reported in the combined analysis (Figure 10). Moderate and strong correlations were found between biomass and NDVI, NormNIR, RVI, DVI, and NG at Site 1 despite the narrow ranges in biomass and lack of significance in N rate (Table 7). The strongest correlations with N concentration ($R^2 = 0.57$ with NG) and N uptake ($R^2 =$

0.83 with DVI) (Figure 11) were found in site 1 (Table 7). Site 2 had moderate correlations between N concentration and NormNIR ($R^2 = 0.50$) and DVI ($R^2 = 0.55$). These differences among sites may be attributed to changing sun angle or other light conditions during photography. Pinter et al. (1983) suggested normalizing sun angle to a standard elevation of 45 degrees to compensate for changes in radiation during the time of sensing. Applying this process to the data in this study might improve correlations, but was not attempted.

CONCLUSIONS

Differences in N concentration of plant tissue at similar biomass were successfully established for a mature stand of bermudagrass. Irrigation did not create differences in the moisture composition of the plant tissue. The combined analysis (irrigation x N rates) revealed significant but weak correlations between N concentration and NIR, NormNIR, DVI, NG, and GNDVI. There was a site x VI interaction for the relationships between N concentration and NG and GNDVI. Strong relationships were found between N concentration and NG and GNDVI for sites 1 and 2. Nitrogen concentration was more strongly estimated by NG ($R^2 = 0.57$) in site 1, and DVI ($R^2 = 0.55$) in site 2; N uptake was strongly estimated by DVI ($R^2 = 0.83$) at sites 1 and 2. Stronger coefficients of determination in site 1 versus 2 and 3 could have been due to changing light conditions associated with the time it took to photograph each replication.

ACKNOWLEDGEMENTS

We thank TJ Holliday, Pete Thompson, Michael Scott, Coy Bridges, and Ray Wilkerson for their field support.

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Table 4. Analysis of variance for the crop response variables for bermudagrass harvested at early heading in September, 2000 in Raleigh, NC.

	Crop Response Variables						
	Tissue Moisture	Biomass	N Concentration	N Uptake	Color	Inflorescence Amount	Soil Moisture
Rep	**	***	NS	***	NS	NS	**
Irrigation	NS	NS	NS	NS	NS	*	*
Rep*Irrigation	*	NS	NS	NS	NS	NS	NS
N	**	NS	***	*	***	NS	NS
N *Irrigation	NS	NS	NS	NS	NS	NS	NS

*, **, and *** indicates significance at the p = 0.05, 0.01, and 0.001 levels, respectively
 NS, not significant

Table 5. Analysis of variance for the spectral variables for bermudagrass harvested at early heading in September, 2000 in Raleigh, NC.

	Spectral Variables									
	Green	Red	NIR	NDVI	Norm NIR	RVI	DVI	NR	NG	GNDVI
Rep	***	***	***	NS	NS	NS	NS	NS	**	NS
Irrigation	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Rep * Irrigation	NS	NS	NS	NS	NS	NS	**	NS	NS	NS
N	NS	NS	**	*	*	*	***	NS	**	**
N * Irrigation	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

*, **, and *** indicates significance at the p = 0.05, 0.01, and 0.001 levels, respectively
NS, not significant

Table 6. Coefficients of determination (R^2) for the relationships between spectral and crop response variables for bermudagrass harvested at early heading in September, 2000 in Raleigh, NC.

Crop Response Variables	Spectral Variables									
	G	R	NIR	NDVI	Norm NIR	RVI	DVI	NR	NG	GNDVI
	----- R^2 -----									
Color†	NS	NS	NS	0.32	0.34	0.31	0.45	0.19	0.09	0.25
Biomass	NS	NS	NS	NS	NS	NS	0.14	NS	NS	NS
N Concentration	NS	NS	0.15	NS	0.15	NS	0.23	NS	0.18	0.19
N Uptake	NS	NS	0.13	0.10	0.15	0.09	0.28	NS	0.10	0.16

NS, not significant

† color scores visually assigned with 1 indicating light, yellowing leaf tissue and 10 indicating dark green leaf tissue.

Table 7. Coefficients of determination (R^2) for the relationships between spectral and crop response variables for each bermudagrass site harvested at early heading in September, 2000 in Raleigh, NC.

	Spectral Variables									
	G	R	NIR	NDVI	NormNIR	RVi	DVI	NR	NG	GNDVI
Site 1	<hr/> R^2 <hr/>									
Biomass	NS	NS	NS	0.70 [†]	0.69 [†]	0.69 [†]	0.69 [†]	0.42	0.69 [†]	0.46
N Concentration	NS	NS	NS	NS	NS	NS	0.31	NS	0.57	0.42
N Uptake	NS	NS	NS	0.37	0.73 [†]	NS	0.83 [†]	NS	0.44	0.62
Site 2										
Biomass	NS	NS	0.33	0.41	0.45	0.41	0.53	NS	0.34	0.43
N Concentration	NS	NS	NS	0.46	0.50	0.44	0.55	0.28	0.38	0.49
N Uptake	NS	NS	0.35	0.47	0.51	0.45	0.60	0.28	0.40	0.50
Site 3										
Biomass	NS	NS	NS	NS	NS	NS	NS	NS	0.28	NS
N Concentration	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
N Uptake	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

[†] indicates a quadratic relationship
NS, not significant

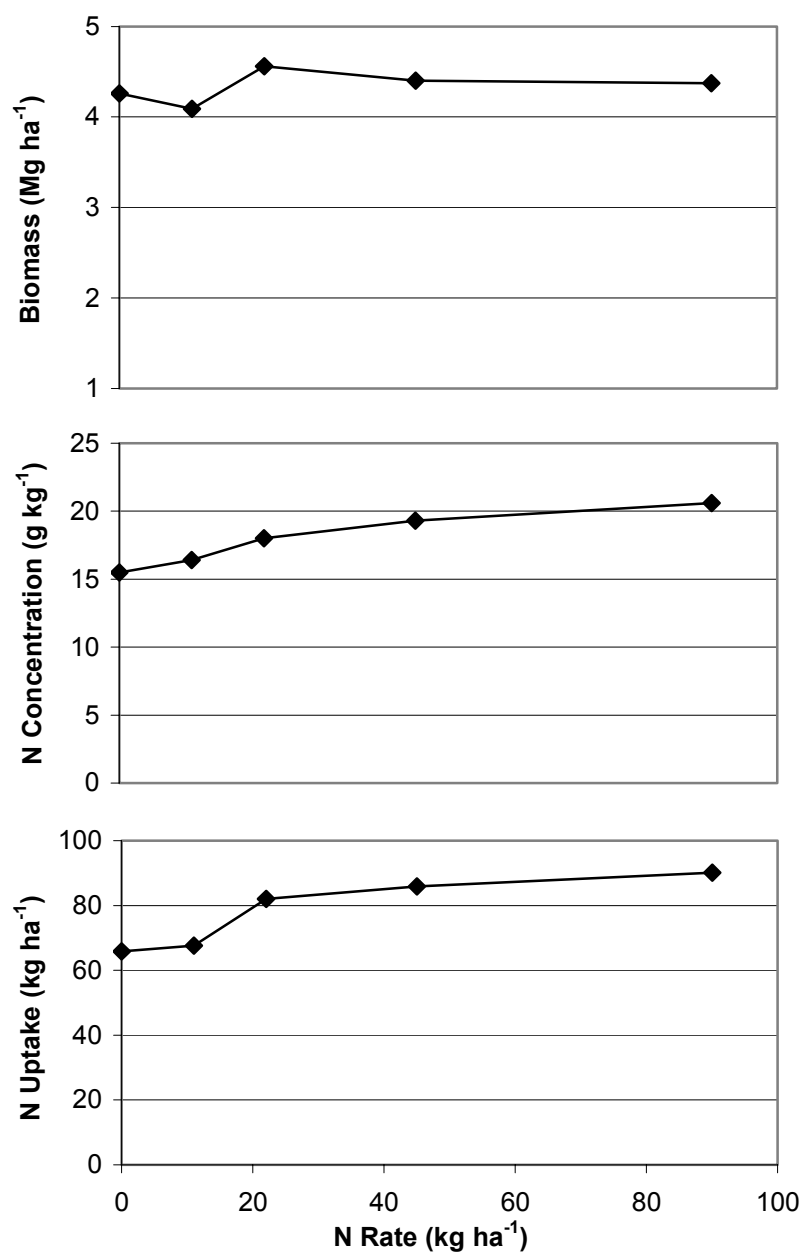


Figure 8. Mean response of biomass, N concentration, and N uptake to N rates in bermudagrass canopies harvested at early heading in September, 2000 in Raleigh, NC.

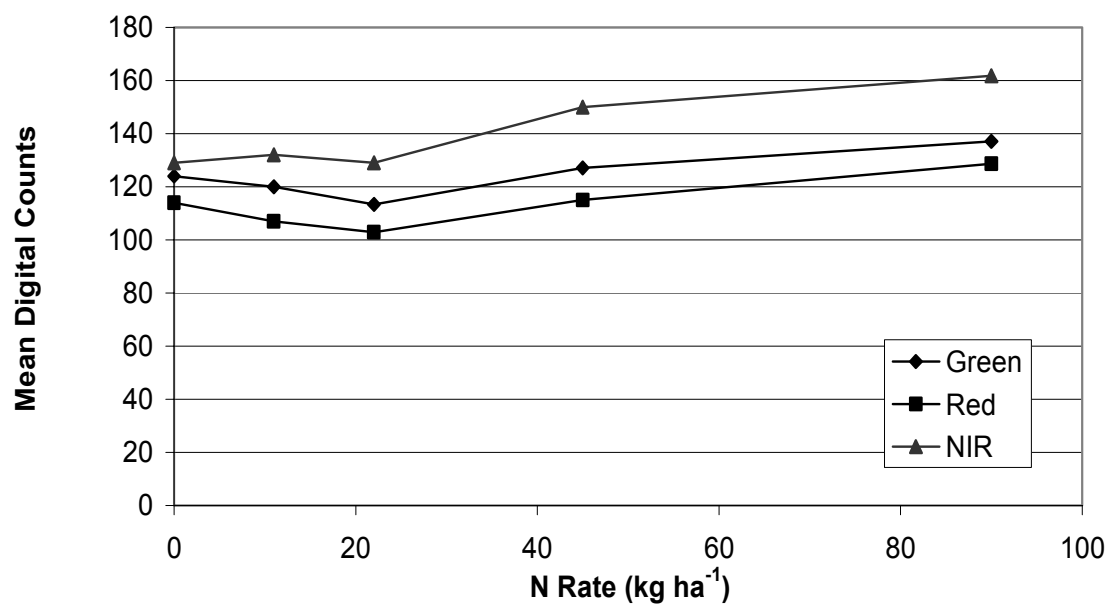
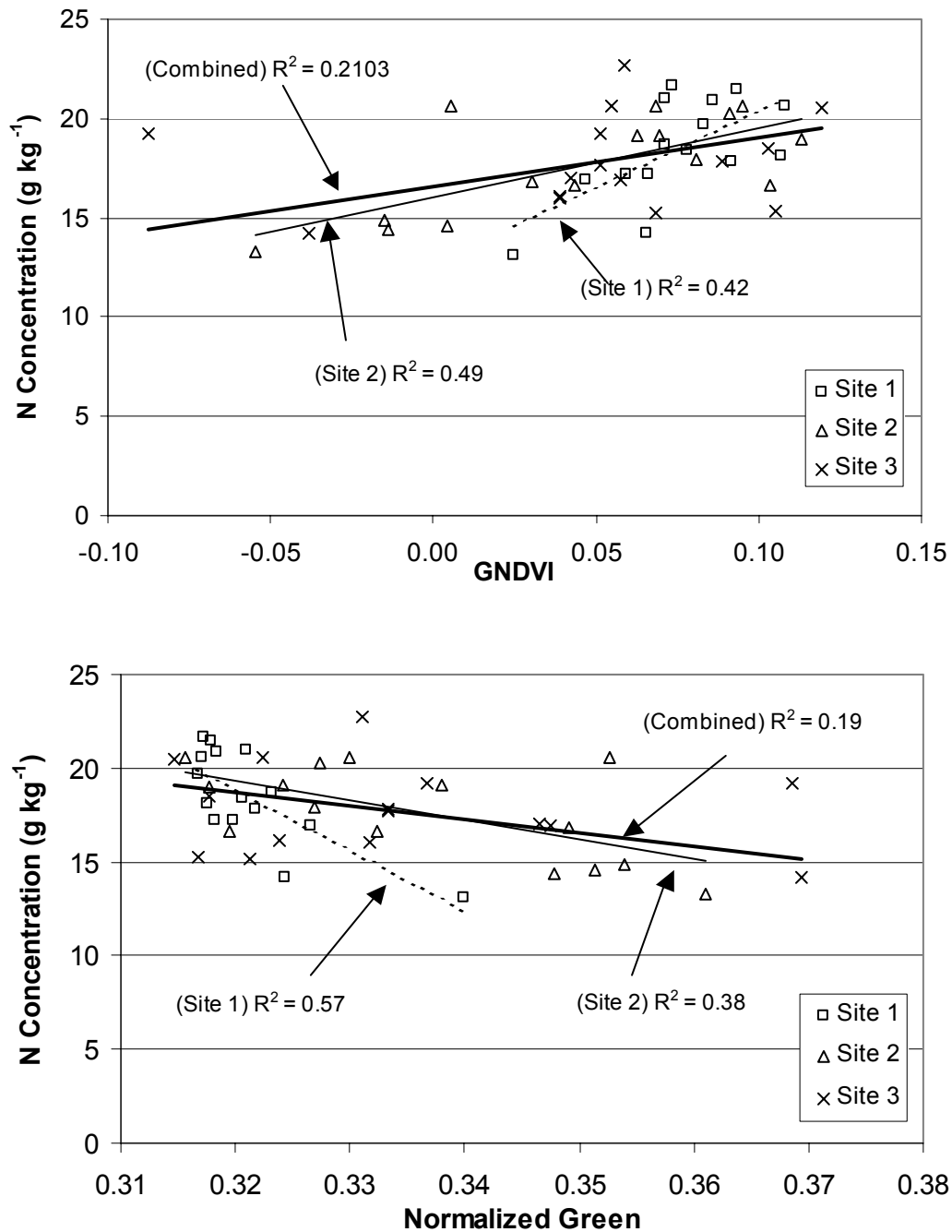
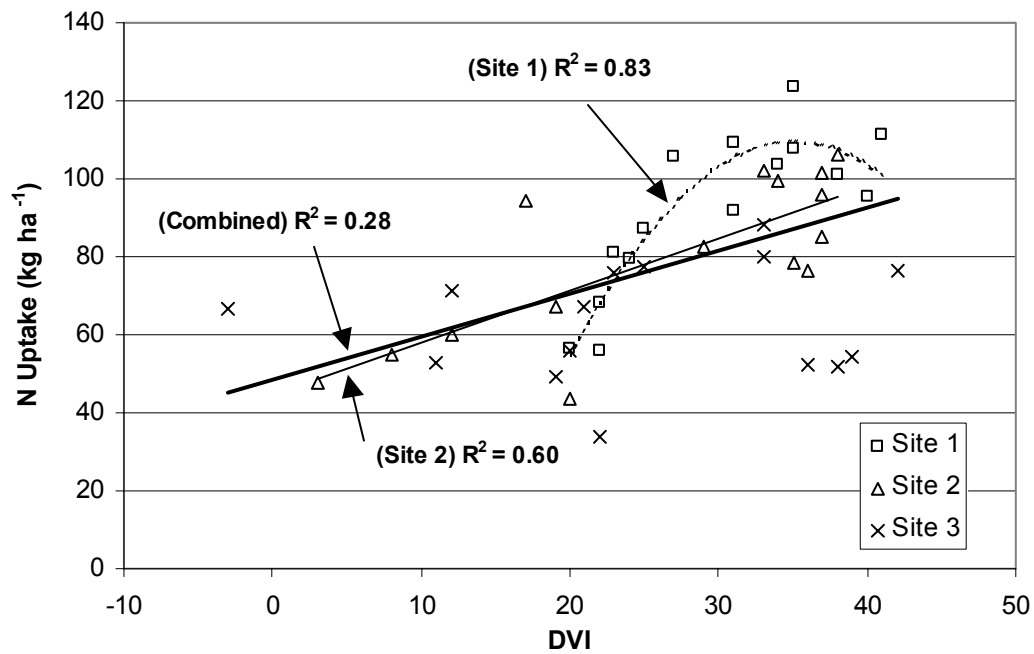


Figure 9. Mean nonnormalized digital counts for near infrared (NIR), red, and green across N rates for bermudagrass canopies harvested at early heading in September, 2000 in Raleigh, NC.



† Relationships were not significant for Site 3

Figure 10. Relationships of N concentration of bermudagrass harvested at early heading in September, 2000 with Green NDVI (GNDVI) and Normalized Green (NG) for each site and all sites (combined)[†] in Raleigh, NC.



† relationships were not significant for Site 3

Figure 11. Relationships between N uptake of bermudagrass harvested at early heading in September, 2000 and DVI for each site and combined sites in Raleigh, NC

**Estimating Biomass and Nitrogen Concentration in Bermudagrass
(*Cynodon dactylon* L.) Using Ground-based False Color Infrared Photography**

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Abbreviations: FCIR, false color infrared; B, blue; G, green; R, red; NIR, near infrared; BG, bermudagrass; NDVI, normalized difference vegetation index; RVI, ratio vegetation index; DVI, difference vegetation index; NormNIR, normalized near infrared; NG, normalized green; NR, normalized red; GNDVI, green normalized difference vegetation index; VI, vegetation index; RYE, realistic yield expectation.

Keywords: biomass, nitrogen, forage, infrared, close range photography, remote sensing, bermudagrass

ABSTRACT

Remotely assessing nitrogen (N) concentration and biomass of grassland canopies can provide land managers and regulatory agencies an ongoing opportunity to quickly determine the grassland inventory that is available for grazing animals, wildlife, or soil protection. The objective of this study was to determine if a vegetation index (VI), NIR, R, or G digital counts derived from false color infrared (FCIR) ground-based photographs could be used to estimate biomass, N concentration, and N uptake in bermudagrass across sites, cuttings, or years. Previously established experiments located throughout the Coastal Plain of North Carolina examining realistic yield expectations of bermudagrass (*Cynodon dactylon* L.) on various soil types provided an opportunity to acquire photographs of multiple sites and cuttings in 2000 and 2001. Each experiment was a randomized complete block design, with four replications of five N rates applied in four applications throughout the summer. False color infrared photographs were taken with a 35-mm camera fitted with a yellow filter and elevated 1.83 m above the ground. Biomass, N concentration, and N uptake were measured at each site to determine if the relationship between a crop response variable and a spectral parameter was consistent among sites, cuttings, and/or years. Relationships between Red, Green, and NIR digital counts and crop response variables for most of the sites were weak, however, normalization generally improved correlations. Moderate or strong correlations between spectral and crop response variables, such as between Green NDVI and N uptake ($R^2 = 0.89$), could be found among all sites and cuttings except one. There were cutting, year, and site interactions with VIs for all three comparisons among sites, cuttings, and years. Despite statistics indicating that harvests were best modeled individually, combined

relationships usually resulted in higher coefficients of determination ($R^2 = 0.66$). Taking into account location, photography method, and environmental conditions, Green NDVI and DVI were best to estimate N concentration across four sites ($R^2 = 0.40$). Standardizing the crop response variables to a relative value might improve correlations with spectral parameters across sites.

INTRODUCTION

Remotely assessing nitrogen (N) concentration and biomass of grassland canopies can provide land managers and regulatory agencies an ongoing opportunity to quickly determine the grassland inventory that is available for grazing animals, wildlife, or soil protection. Unlike instruments that measure reflectance directly, false color infrared (FCIR) photography captures all the light in a given scene regardless of source (e.g. reflectance, transmittance, and scatter) onto a film emulsion, which can be scanned and digitized so that the brightness of each pixel within the photograph is quantitatively indicative of the light emanating from a given area on the landscape (Blackmer and Schepers, 1996). Researchers have used FCIR aerial photography to estimate ground cover in row crops, detect winter injury in alfalfa (*Medicago sativa* L.), and predict forage biomass and N content (Gerbermann et al., 1976; Wallen et al., 1977; Ryan et al., 1989). Ground-based FCIR photography has the potential to minimize variability associated with atmospheric conditions, which affects aerial or satellite photography (Scharf and Lory, 2000).

Previous studies by the authors examined the utility of FCIR photography in estimating biomass and N concentration in bermudagrass from both aerial and ground-based platforms for individual sites. The objective of this study was to determine if a vegetation index (VI), NIR, R, or G digital counts derived from FCIR ground-based photographs could be used to estimate biomass and N concentration in bermudagrass across sites, harvests, and years.

MATERIALS AND METHODS

Data were collected from experiments in four locations in the southern Coastal Plain of North Carolina: two locations in Sampson County (1S and 2S), and one each in Bladen County (BL) and Duplin County (DU). Each location was part of a study examining realistic yield expectations (RYE) of bermudagrass (*Cynodon dactylon* L. 'Coastal') on several soils (Table 8). Each site was a randomized complete block design, with four replications of five N rates applied in four target applications (April = 25%, June = 35%, July = 25%, August = 15%). Nitrogen rates were chosen to represent various proportions (0, 75%, 100%, 125% and 200%) of the recommended rate for bermudagrass on similar soil types (Table 8) and applied in the form of ammonium nitrate (NH₃NO₄). Nitrogen rates for each site were: 0, 231, 309, 386, and 617 kg ha⁻¹ yr⁻¹ for BL and DU; 0, 187, 253, 314, and 507 kg ha⁻¹ yr⁻¹ for 1S; and 0, 147, 196, 247, and 395 kg ha⁻¹ yr⁻¹ for 2S.

To capture as much variation in the plots as possible, each harvest area was stratified into three sections and a photograph was taken in each section with a 35-mm SLR manual

camera (Model OM-1, Olympus America Inc., Melville, NY) elevated 1.83 m above the ground and leveled for a nadir view over a ground area of 0.25 m². Kodak Ektachrome Color Infrared EIR film (Eastman Kodak Co., Rochester, NY), which responds to Blue, G, R, and NIR wavelengths, was combined with a yellow filter (Kodak Wratten Gelatin Filter No.12) to absorb blue radiation. Plots were photographed over a two hour period, approximately 12 hours before harvest.

Leaf spot (*Bipolaris cynodontis*) was observed at Site 1S and the 3rd cutting at Site 2S in August, 2000. A score of 1 was assigned for low infections of leaf spot and 10 for high. Forage was harvested (3rd cutting) in August, 2000 at sites DU, BL, 1S, and 2S. Additionally, forage was harvested from site 2S in July 2000 and 2001 (2nd cuttings). Above ground (> 5 cm) biomass was harvested using a Carter flail-type forage harvester (Carter Harvesters, Brookston, IN) from a 4 m² area and weighed immediately. Subsamples from each harvest area were weighed, dried in a forced-air oven at 65 °C for 24 to 48 hours, reweighed to determine tissue moisture, ground to 1 mm, thoroughly mixed, and analyzed for N concentration using a Perkin Elmer 2400 CHN Elemental Analyzer (Perkin Elmer Corp., Norwalk, CT.). Biomass was calculated as the aboveground dry matter yield. Nitrogen uptake was determined by multiplying N concentration by biomass.

The film was processed onto a false color positive slide, which was digitized (.TIFF files) using a slide scanner (Konica Q-Scan, Konica Corp., Mahwah, NJ.) with a resolution of 47 pixels per mm (1000 dpi). The resulting image consisted of a 24-bit RGB image (8 bit

Red, 8 bit Green, 8 bit Blue) with each primary color represented by a digital count within the range of 0 to 255. Pixel brightness values for the three color bands represented in the film (R, G, and NIR) were extracted using ERDAS Imagine Version 8.3.1 (ERDAS, 1997). Pixel values from each of the photographed sections of the harvest area were averaged to generate a single value per plot for each color band. In addition to the digital values in each band, a number of ratios were derived (Table 1) and used to develop relationships with crop response variables (N concentration, biomass, and N uptake).

The data were analyzed using analysis of variance and simple regression in SAS Version 8 (SAS, Cary, NC). Differences referred to hereafter imply significance at $p \leq 0.05$. For each harvest, regression and correlation analyses were performed to assess the relationships between spectral and crop response variables. A sequence of linear versus quadratic models was determined using Proc GLM. Proc REG was used to determine Pearson correlation coefficients when a linear model was adequate. Based on the harvest data, comparisons were made between the 2nd and 3rd cuttings (July and August, 2000) from the same site (2S), 2nd cuttings for two years (July 2000 and 2001) at the same site (2S), and the 3rd cutting (August, 2000) from four different sites (DU, BL, 1S, and 2S). PROC MIXED was used to test for differences between harvests within these comparisons. Crop response variables were analyzed and presented graphically as the dependent variable in order to test the spectral indices as estimators of those variables. For the purposes of this paper, correlations were defined as strong ($R^2 > 0.7$), moderate ($0.5 < R^2 < 0.7$), and weak ($R^2 < 0.5$).

RESULTS AND DISCUSSION

Spectral relationships with biomass, N concentration, and N uptake at each site

Site DU. The third cutting from this site was photographed in August, 2000. Values for the crop response variables generally increased with N rate (Figure 12). The only significant relationships between crop response variables and raw digital counts were between NIR and biomass and N uptake (Table 9). Normalizing the raw digital counts generally resulted in stronger relationships between crop response variables and VIs. Biomass was best estimated by NIR digital counts ($R^2 = 0.41$). Moderate linear relationships were found for DVI with N concentration ($R^2 = 0.63$) and with N uptake ($R^2 = 0.54$).

Site BL. The third cutting from this site was photographed in August, 2000. Biomass and N uptake increased until 300 kg N ha⁻¹ and then plateaued, while N concentration increased across N rates (Figure 12). There were no significant relationships between raw digital counts and crop response variables (Table 9). Furthermore, biomass was not correlated with any of the VIs. A moderate quadratic relationship was found between N concentration and NormNIR ($R^2 = 0.54$). Nitrogen uptake was best estimated by DVI ($R^2 = 0.35$). All plots at this site were infested by crabgrass (*Digitaria sanguinalis* L.) which ranged from 2 to 35 % (Mean = 11%) of the stands. Previous studies by the author

found lower correlations between spectral and crop response variables in crabgrass canopies versus bermudagrass canopies (Morgan, 2002).

Site 1S. The 3rd cutting from this site was photographed in August, 2000. Leaf spot infestation was observed in the canopy, with visual scores ranging from 2 to 8 (mean = 5.5). Biomass and N uptake were relatively low across N rates (Figure 12), plateauing beyond the first N increment, while N concentration increased with increasing N. For the raw digital counts, weak relationships were found between R digital counts and biomass and N uptake, and between G digital counts and N uptake (Table 9). Coefficients of determination (R^2) improved dramatically when VIs were used to estimate crop response variables. Strong relationships were found between all the VIs and biomass ($R^2 = 0.77$ to 0.87) and N uptake ($R^2 = 0.84$ to 0.89), while moderate relationships were found between all the VIs and N concentration ($R^2 = 0.53$ to 0.69) except for NG, which had a strong correlation ($R^2 = 0.83$).

Site 2S. The 2nd and 3rd harvests were photographed in July and August of 2000 respectively, as well as the 2nd harvest the following year in July, 2001. Leaf spot was evaluated at the August 2000 harvest with infestation levels ranging from 2 to 6 (mean = 4.6). Biomass, N concentration, and N uptake in July and August, 2000 increased with N rate (Figure 13). Among raw digital counts in July 2000, weak to moderate correlations were found between crop response variables and NIR digital counts, which was the best estimator of N concentration ($R^2 = 0.45$) (Table 9). Normalizing the raw digital counts did not generally improve correlations, however, DVI was the best estimator of biomass

($R^2 = 0.67$) and N uptake ($R^2 = 0.59$). Among raw digital counts in August 2000, a moderate relationship was found between R and N concentration ($R^2 = 0.55$) and weaker relationships were found between R and N uptake and G and N concentration. With the exception of NG, normalizing the raw digital counts strengthened relationships for most of the crop response variables. Biomass was best estimated by NormNIR ($R^2 = 0.59$), while N concentration was best estimated by NR ($R^2 = 0.73$). N uptake was equally correlated with NDVI, NormNIR, and RVI.

Biomass and N uptake for the July, 2001 harvest from this site increased with N rate but were half those observed in the 2nd cutting from the previous year, while N concentration values were similar (Figure 14). Weak correlations were found between R digital counts and biomass and N uptake, while G and NIR digital counts were not significantly correlated with any of the crop response variables (Table 9). Biomass and N uptake were best estimated by NDVI and NR, while N concentration was best estimated by a quadratic relationship with RVI. A brief rain shower occurred in the middle of photography and subsequent light conditions varied considerably from sunny to cloudy. Precipitation on the day of sensing has been reported to cause decreases in canopy reflectance and could be a reason for the lower coefficients of determination for NDVI in Figure 15 (Daughtry et al., 1980). Despite postponing photography for an hour while the canopy "dried", it is likely that water droplets were present on the leaf blades for at least half of the plots photographed. With only 1.83 m between the canopy and the camera, effects of dew and raindrops on the leaves could be considerable. Conversely, Mayhew

et al. (1984) reported no effect of moisture on RVI readings from a mixed sward canopy sensed with a spectrophotometer after a rain (height above ground = 1.25 m).

The effect, if any, of leafspot fungus at these sites is difficult to isolate. Biomass was lower for the 3rd versus the 2nd cutting at site 2S, but could be the result of multiple environmental factors. Discoloration of leaf tissue may have altered plant pigments enough to affect G and R raw digital counts. Knipling (1970) reported increases in visible reflectance when chlorophyll metabolism was disrupted. Gausman (1974) observed that internal discoloration of leaves resulting in a brown pigmentation caused decreases in NIR reflectance. Knipling (1970) observed a greater decrease in NIR versus visible reflectance for canopies that had been stressed (loss of leaves, changes in orientation, or overall suppression of growth) due to fewer leaf layers and increased soil exposure. Sinclair (1968) suggested that the increase in reflectance from leaves that were stressed by disease was confounded with effects of dehydration, which similarly changes internal leaf structure and depresses NIR reflectance. Given the close proximity of the camera to the canopy, leaf effects may be more important than when photographed from an aerial platform.

Spectral relationships with crop response variables for two cuttings at the same site in 2000

There was a cutting x VI interaction for all the relationships between VIs and biomass and N uptake, and for the relationships of N concentration with NDVI, RVI, and GNDVI (Table 10, Figure 15). Despite the interaction for biomass and N uptake with NDVI in

Figure 15, the correlation for the combined cuttings was stronger than the individual correlations for each cutting. This trend was not evident for the relationship between cuttings and N concentration, whereby the 3rd cutting was more strongly correlated with NDVI. These trends were consistent for all the relationships between biomass, N concentration, and N uptake and the VIs where there was an interaction except for the relationship between GNDVI and N uptake, which resulted in a stronger correlation for the 2nd cutting versus the 3rd or combined cuttings.

Spectral relationships with crop response variables for two similar cuttings at the same site in 2000 and 2001

Comparisons were made between the 2nd cuttings at site 2S in 2000 and 2001 to see if correlations were similar across years. Crop response to N at these harvests was similar to the previous comparison of the 2nd and 3rd cutting in 2000 (Figures 13 and 14).

Correlations between crop response variables and VIs were affected by year (Table 11) for all but three relationships and had similar interactions as were noticed with the comparison between the 2nd and 3rd cuttings in 2000. The relationships of biomass and N concentration with NDVI (Figure 16) showed a larger range in crop response values as compared to the 2nd and 3rd cuttings in 2000, but in different ranges of NDVI. The strengths of the relationships for each year were similar, indicating that the same amount of variability was being accounted for each year. This tendency was less for N concentration than biomass.

**Spectral relationships with crop response variables for the 3rd cuttings of
bermudagrass in August, 2000 at four different sites**

Biomass and N uptake across N rates for the August 2000 cuttings at sites DU and BL were much greater than at sites 1S and 2S (Figure 12). Nitrogen concentration tended to increase with N rate similarly at all sites except site DU where it plateaued at the higher rates. The lower yields for sites 1S and 2S may have been due, in part, to leafspot, which was present in both cuttings. Alternatively, the fine sandy soils (Table 8) at these sites could be lower yielding. Realistic yield expectations for bermudagrass on these soils were 3.5 tons ac⁻¹ versus 5.5 tons ac⁻¹ for sites BL and DU. Coefficients of determination (R^2) for the relationships between spectral and crop response variables were generally lower than those observed in the previous two comparisons (Table 12 versus Tables 10 and 11). Site interactions affected all the relationships between crop response variable and VIs except among N concentration, N uptake, DVI, and GNDVI. Individual site relationships (Table 9) were stronger than the combined site analysis (Table 12) for estimating biomass and N uptake but weaker for estimating N concentration. Green NDVI and DVI were able to estimate N concentration ($R^2 = 0.40$) despite variability associated with location, photography method, and environmental conditions (Figure 17). Stronger correlations may result by normalizing the data to a relative scale, such as dividing each crop response value by the mean.

CONCLUSIONS

Relationships among raw digital counts and crop response variables for most of the sites were weak, however, normalization generally improved correlations. Moderate or strong correlations between spectral and crop response variables could be found among all sites and cuttings except for July, 2001 at site 2S. There were cutting, year, and site interactions with VIs for all three comparisons. Despite statistics indicating that harvests were best modeled individually, the combined relationships usually resulted in higher coefficients of determination, indicating that a common relationship may be more useful in estimating biomass, N concentration, and N uptake in bermudagrass.

Standardizing the crop response variables to a relative value has been found to improve correlations with spectral parameters (Blackmer et al., 1996; Flowers et al., 2001). This could be done by calculating crop response variables as a percent of maximum yield, dividing individual values by the mean, or by subtracting the mean from each value and dividing by the variance. Applying one of these procedures could improve the robustness of Green NDVI as an estimator of biomass, N concentration, and N uptake.

ACKNOWLEDGEMENTS

We thank Michael Scott, TJ Holliday, and Pete Thompson for their field support.

Funding was provided in part by the North Carolina Agriculture Foundation and the North Carolina Department of Environmental Resources.

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Table 8. Soil description, surface soil test results (0 to 10 cm), and realistic yield expectations (RYE) for bermudagrass canopies located in Duplin (DU), Bladen (BL), and Sampson (1S and 2S) counties in North Carolina.

Site	Soil Type	Taxonomy	Soil Test Information			RYE
			pH (H ₂ O)	P [‡]	K [‡]	
				—————Kg ha ⁻¹ —————		Mg ha ⁻¹
DU	Kenansville	Loamy, siliceous, subactive, thermic Arenic Paleudults	5.1	350	17	12
BL	Butters	Coarse-loamy, siliceous, semiactive, thermic Typic Paleudults	5.3	406	25	12
1S	Cainhoy	Fine sandy, thermic, coated Typic Quartzipsamments	6.3	97	34	7.8
2S	Cainhoy	Fine sandy, thermic, coated Typic Quartzipsamments	5.8	120	38	7.8

‡ Mehlich, 1984

Table 9. Coefficients of determination (R^2) for relationships between raw digital counts and vegetation indices (VIs) with biomass, N concentration, and N uptake for six cuttings at four locations (Coastal Plain, North Carolina) between July 2000 and 2001.

Crop Response Variables	Spectral Variables									
	G	R	NIR	NDVI	Norm NIR	RVI	DVI	NR	NG	GNDVI
Site DU (August 2000)	R^2									
Biomass	NS	NS	0.41 [†]	0.26 [†]	0.31 [†]	NS	0.20	NS	0.26 [†]	0.37 [†]
N Concentration	NS	NS	NS	0.39	0.49	0.37	0.63	0.28	0.22	0.53
N Uptake	NS	NS	0.24	0.41 [†]	0.30	0.40 [†]	0.54	0.36 [†]	NS	0.34
Site BL (August, 2000)										
Biomass	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
N Concentration	NS	NS	NS	0.38	0.54 [†]	0.34	0.37	0.33	NS	0.48 [†]
N Uptake	NS	NS	NS	0.28	0.26	0.26	0.35	0.27	NS	0.30
Site 1S (August, 2000)										
Biomass	NS	0.31	NS	0.87 [†]	0.87 [†]	0.87 [†]	0.87	0.86 [†]	0.77	0.83
N Concentration	NS	NS	NS	0.53	0.60	0.63 [†]	0.60	0.60 [†]	0.83	0.69
N Uptake	0.25	0.32	NS	0.86 [†]	0.87 [†]	0.86 [†]	0.87	0.84 [†]	0.89	0.89
Site 2S (July, 2000)										
Biomass	NS	NS	0.44	0.43 [†]	0.40 [†]	0.39 [†]	0.67	0.30	NS	NS
N Concentration	NS	NS	0.45	0.26 [†]	0.24 [†]	0.26 [†]	0.29	0.27 [†]	NS	NS
N Uptake	NS	NS	0.56	0.39 [†]	0.36 [†]	0.36 [†]	0.59	0.41 [†]	NS	0.32 [†]
(August, 2000)										
Biomass	NS	NS	NS	0.54 [†]	0.59 [†]	0.55 [†]	0.55 [†]	0.28	NS	0.45 [†]
N Concentration	0.37	0.55	NS	0.69	0.61	0.67	0.67	0.73	NS	0.41
N Uptake	NS	0.28	NS	0.56 [†]	0.56 [†]	0.57 [†]	0.42	0.41	NS	0.28
(July, 2001)										
Biomass	NS	0.21	NS	0.47	0.43	0.44	0.45	0.47	NS	0.33
N Concentration	NS	NS	NS	0.36	0.32	0.47 [†]	0.37	0.37	NS	0.24
N Uptake	NS	0.25	NS	0.45	0.40	0.42	0.42	0.46	NS	0.29

NS, not significant

[†] indicates a quadratic relationship

Table 10. Coefficients of determination (R^2) for relationships between raw digital counts and vegetation indices (VIs) with biomass, N concentration, and N uptake for bermudagrass harvested in July (2nd cutting) and August (3rd cutting), 2000 at Site 2S.

Crop Response Variables	Spectral Variables									
	G	R	NIR	NDVI	NormNIR	RVI	DVI	NR	NG	GNDVI
	R^2									
Biomass	0.08	0.39	NS	0.66^{†§}	0.61^{†§}	0.67^{†§}	0.78[§]	0.63[§]	0.20[§]	0.32[§]
N Concentration	NS	0.21[†]	NS	0.25^{†§}	0.28[†]	0.17^{†§}	0.32[†]	0.21[†]	NS	0.26^{†§}
N Uptake	NS	0.29	NS	0.61^{†§}	0.56^{†§}	0.61^{†§}	0.72[§]	0.60^{†§}	0.19[§]	0.25^{†§}

[†] indicates a quadratic relationship
 NS, not significant
[§] harvest x VI interaction

Table 11. Coefficients of determination (R^2) for relationships between raw digital counts and vegetation indices (VIs) with biomass, N concentration, and N uptake for bermudagrass harvested in July (2nd cutting) of 2000 and 2001 at Site 2S.

Crop Response Variables	Spectral Variables									
	G	R	NIR	NDVI	NormNIR	RVI	DVI	NR	NG	GNDVI
	R^2									
Biomass	0.23	0.37	NS	0.53[§]	0.50[§]	0.56^{†§}	0.48[§]	0.55[§]	0.10	0.43[§]
N Concentration	NS	NS	NS	0.39^{†§}	0.34^{†§}	0.40^{†§}	0.36	0.42^{†§}	NS	0.15
N Uptake	0.15	0.30	NS	0.46[§]	0.42[§]	0.51^{†§}	0.46[§]	0.48^{†§}	NS	0.36[§]

[†]indicates a quadratic relationship

NS, not significant

[§] year x VI interaction

Table 12. Coefficients of determination (R^2) for relationships between raw digital counts and vegetation indices (VIs) with biomass, N concentration, and N uptake for bermudagrass harvested in August (3rd cutting) of 2000 at Sites DU, BL, 1S, and 2S.

Crop Response Variables	Spectral Variables									
	G	R	NIR	NDVI	NormNIR	RVI	DVI	NR	NG	GNDVI
	R^2									
Biomass	NS	0.18 [†]	NS	0.31 ^{†§}	0.27 ^{†§}	0.31 ^{†§}	0.26 [§]	0.34 ^{†§}	NS	0.19 ^{†§}
N Concentration	0.17	0.24	NS	0.47 ^{†§}	0.47 ^{†§}	0.48 ^{†§}	0.41 [†]	0.45 ^{†§}	0.09 [§]	0.40 [†]
N Uptake	0.06	0.16	NS	0.36 ^{†§}	0.36 ^{†§}	0.36 ^{†§}	0.30	0.39 ^{†§}	NS	0.23 [†]

[†] indicates a quadratic relationship
 NS, not significant
 § site x VI interaction

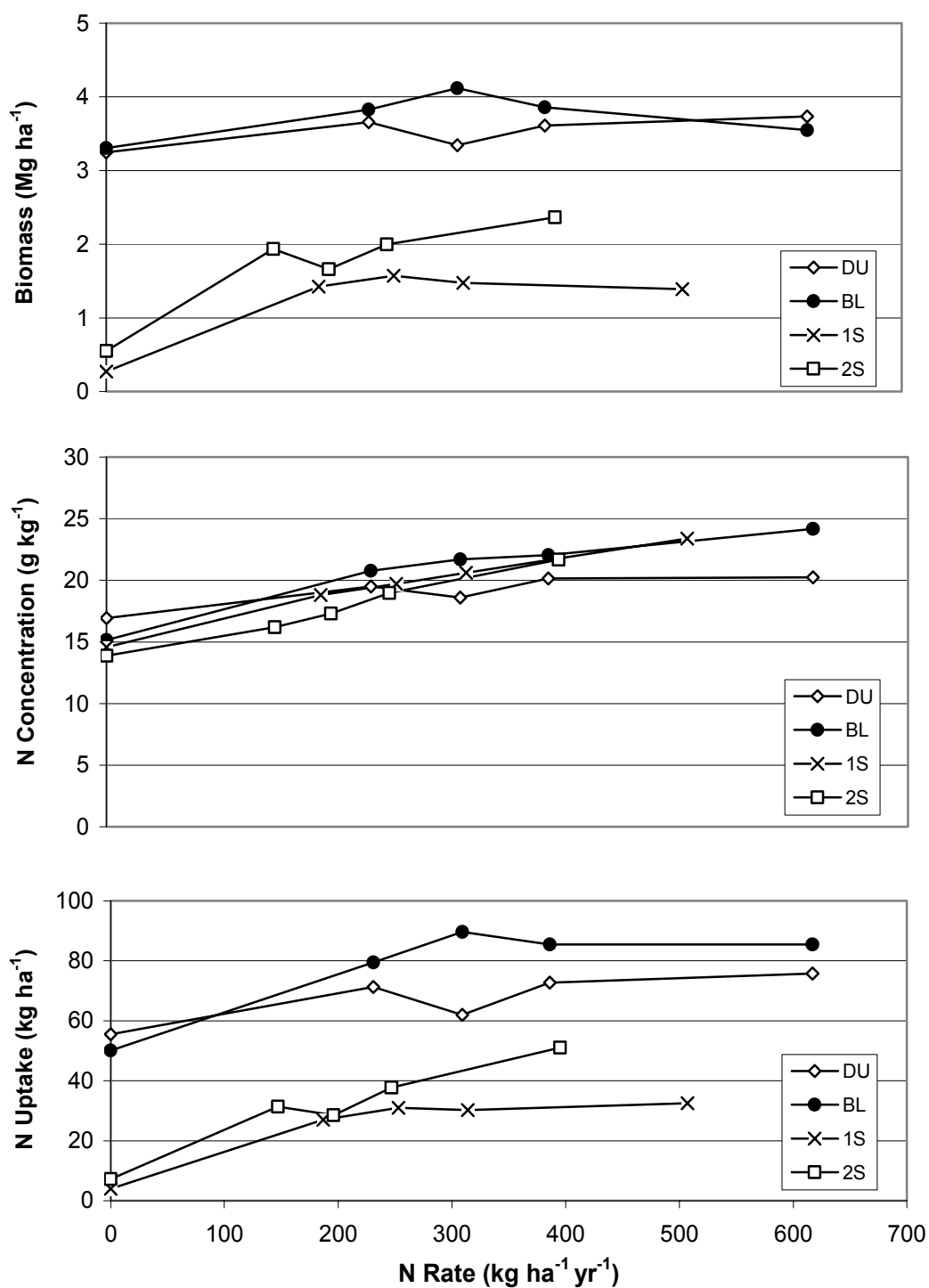


Figure 12. Influence of N rates on biomass, N concentration, and N uptake of bermudagrass harvested in August (3rd cutting), 2000 from four sites.

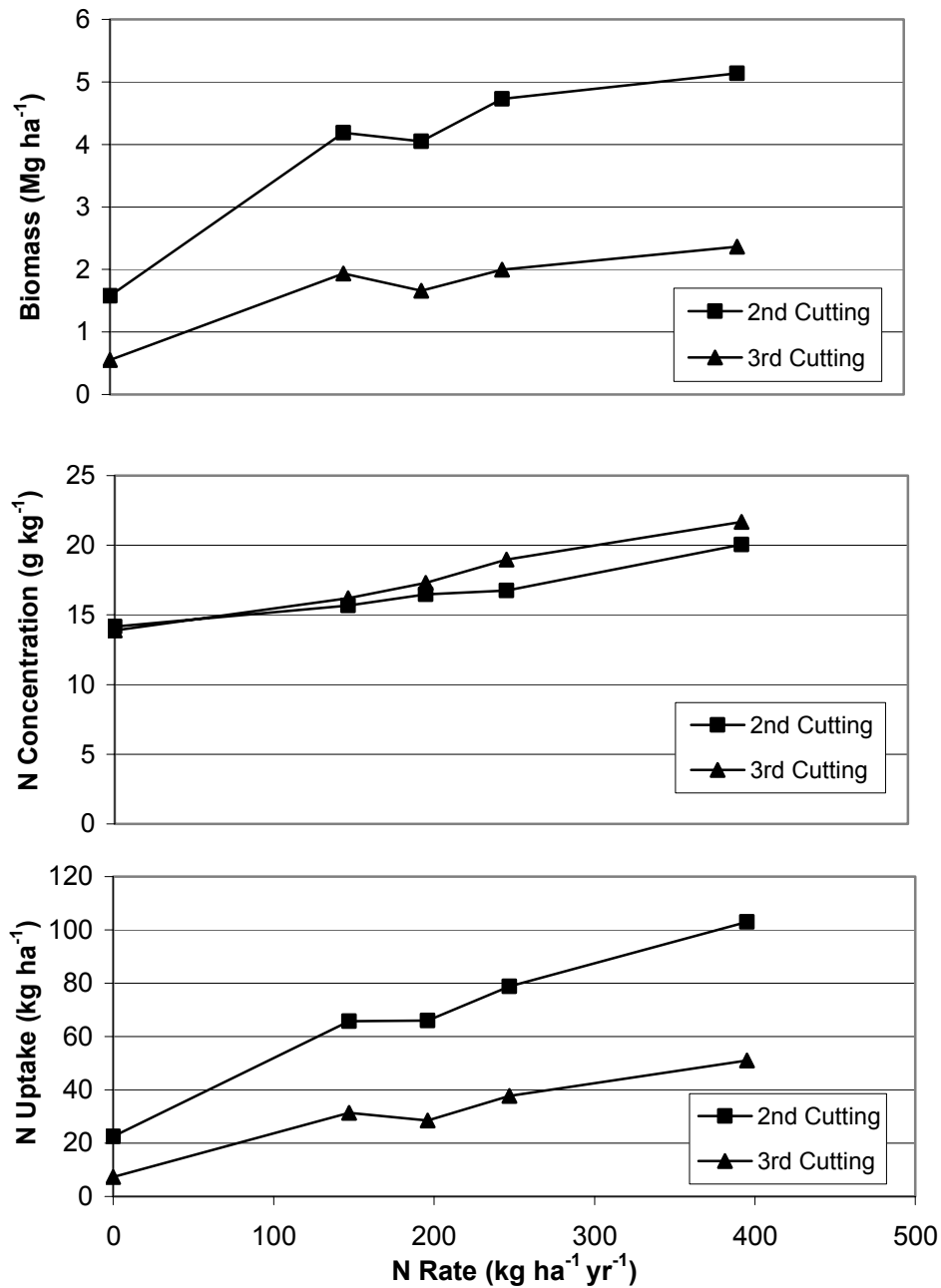


Figure 13. Influence of N rates on biomass, N concentration, and N uptake of bermudagrass harvested in July (2nd cutting) and August (3rd cutting), 2000 at site 2S.

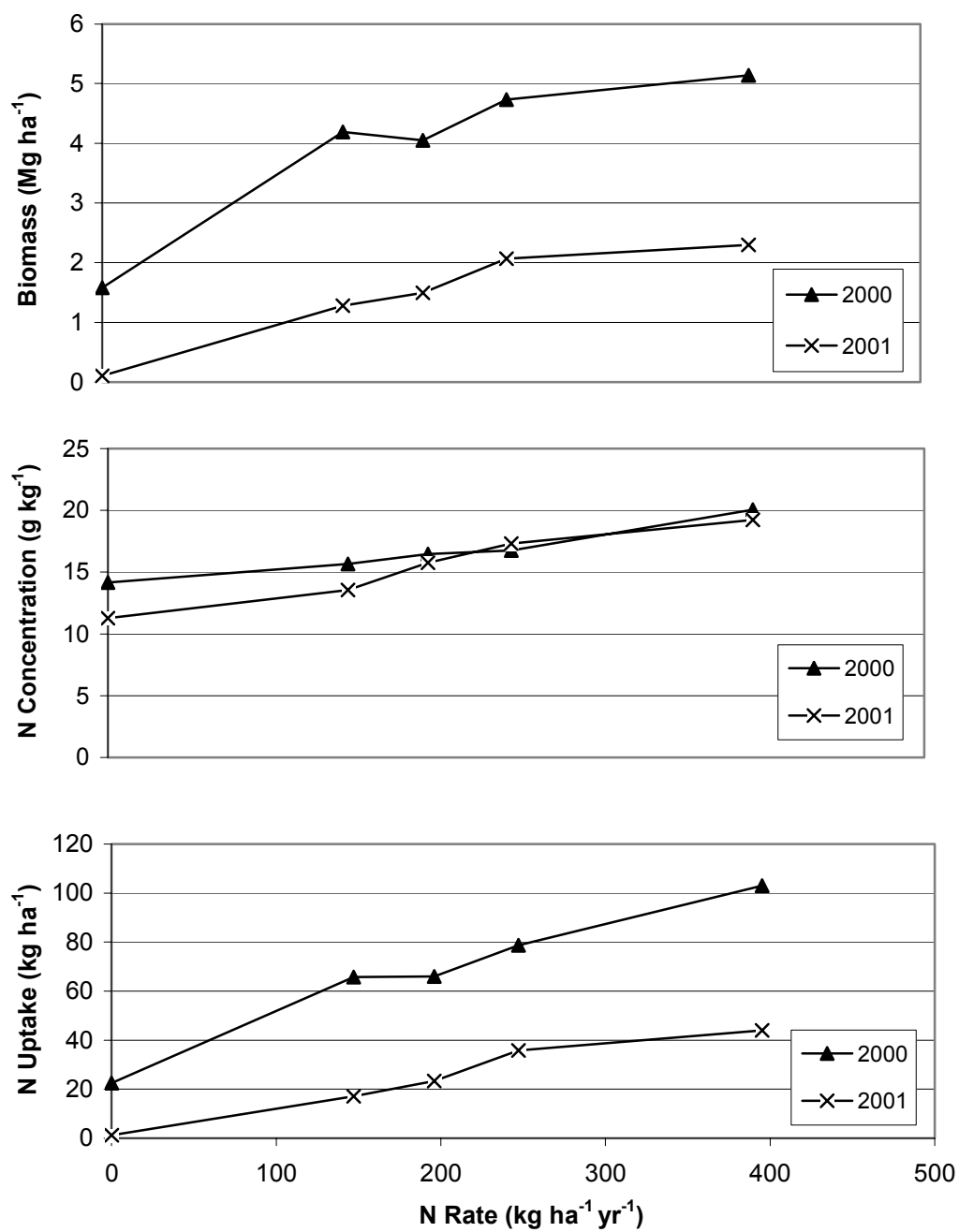


Figure 14. Influence of N rates on biomass, N concentration, and N uptake of bermudagrass harvested in July (2nd cutting) of 2000 and 2001 at Site 2S.

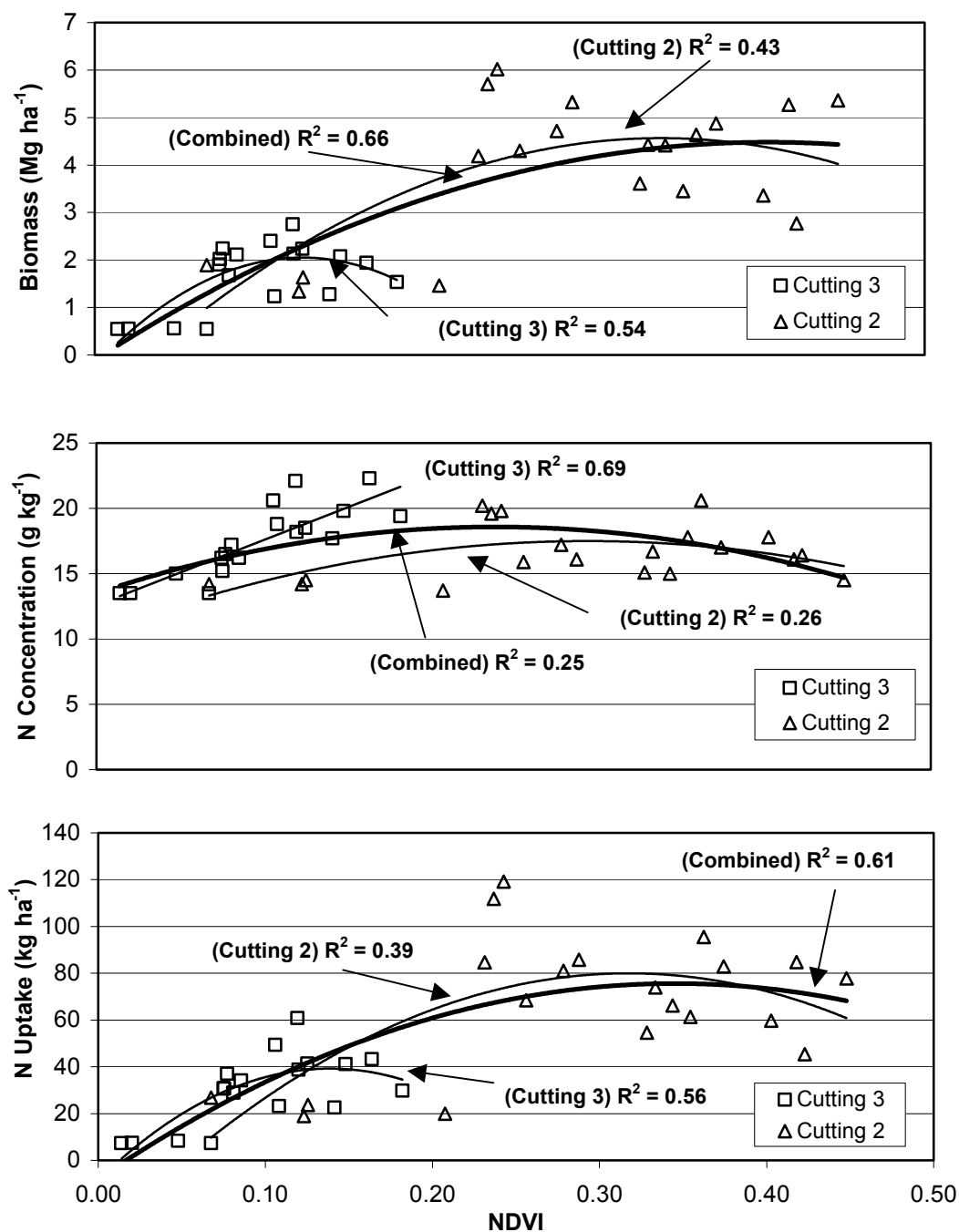


Figure 15. The relationship between NDVI and biomass, N concentration, and N uptake of bermudagrass harvested in July (2nd cutting) and August (3rd cutting), 2000 at Site 2S.

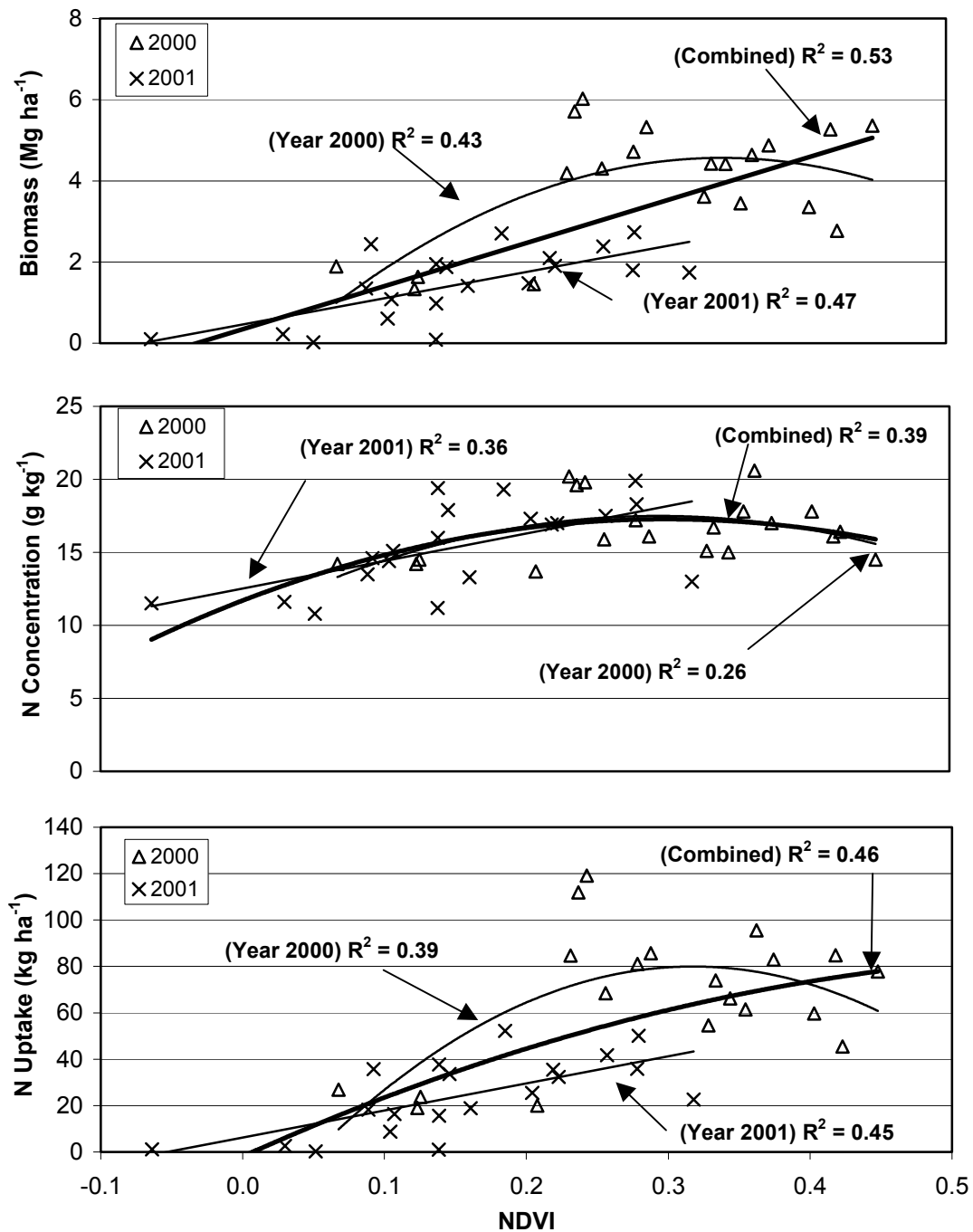


Figure 16. The relationships of NDVI with biomass, N concentration, and N uptake of bermudagrass harvested in July (2nd cutting) of 2000 and 2001 at Site 2S.

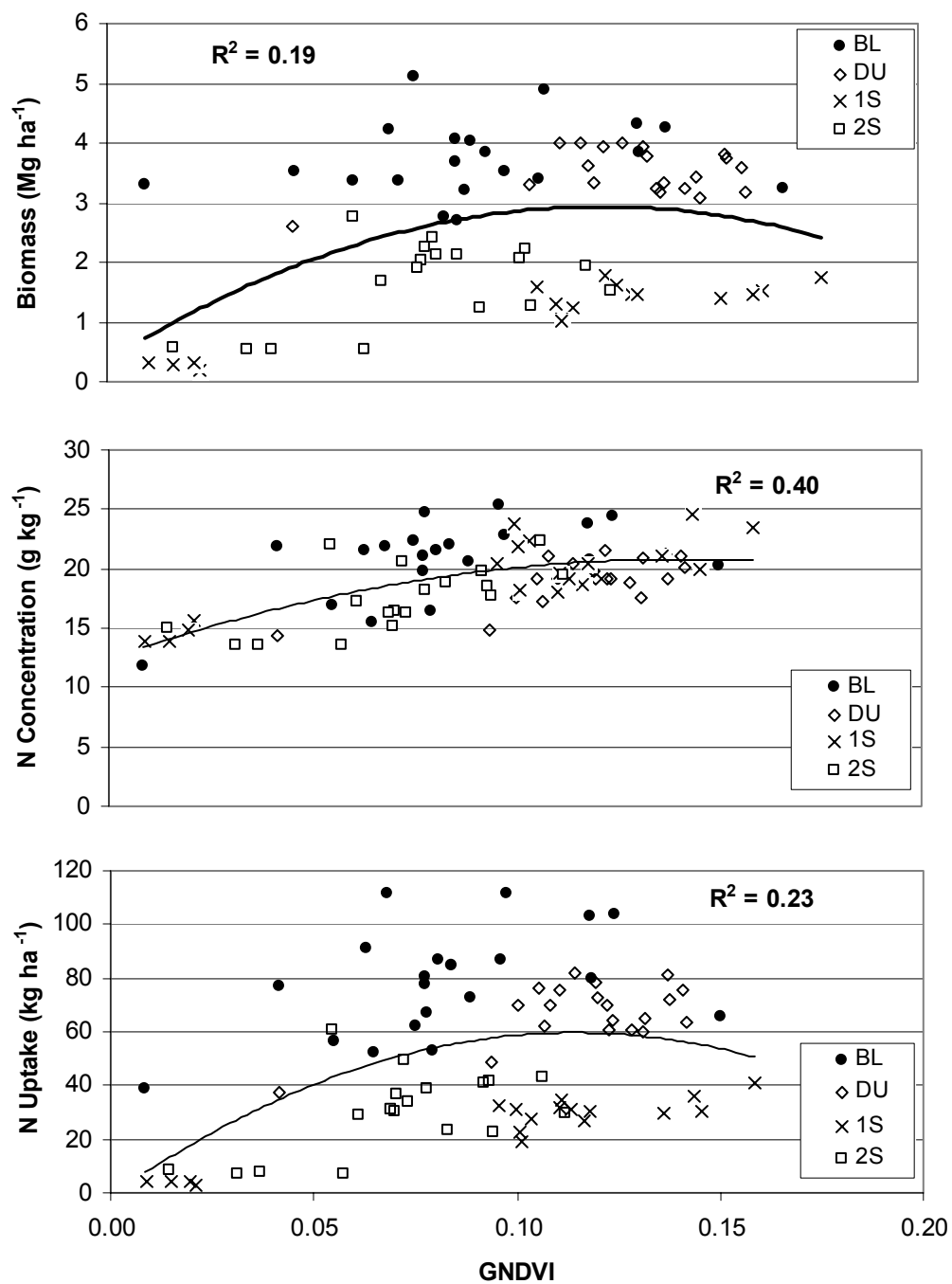


Figure 17. The relationship between Green NDVI and biomass, N concentration, and N uptake of bermudagrass harvested in August (3rd cutting), 2000 at four different sites.

THESIS CONCLUSIONS

This research indicates that false color infrared aerial and ground photography has the potential to aid in the estimation of forage canopy biomass, N concentration, and N uptake. Relationships between forage biomass, N concentration, N uptake with raw digital counts, and vegetation indices (VIs) obtained from false color infrared (FCIR) aerial photography were generally different among forage species. The strongest correlations (R^2) for these relationships were found in bermudagrass (BG) canopies versus crabgrass (CG) and volunteer warm season (VWS) canopies. Normalized raw digital counts were generally stronger estimators of biomass, N concentration, and N uptake than raw digital counts with the exception of the relationship between biomass and NIR digital counts. Biomass was best estimated by NIR digital counts in BG ($R^2 = 0.82$), NDVI in CG ($R^2 = 0.54$), and NormNIR in VWS ($R^2 = 0.86$). Nitrogen concentration was best estimated by NDVI in BG ($R^2 = 0.62$), NIR digital counts in CG ($R^2 = 0.56$), and G digital counts in VWS ($R^2 = 0.63$). Green NDVI was a consistently strong estimator ($R^2 > 0.76$) of N uptake for all forage canopies and was unaffected by N source. Effects of N source were greater in VWS canopies, but this may have due to the exclusion of 20% of the plots fertilized with NH_4NO_3 due to foliar injury. Green NDVI appeared to be a robust indicator of N uptake among different forage canopies and N sources. Estimating N uptake aurally throughout eastern NC could provide regulatory agencies and farmers with a quick and accurate resource for determining the N efficiency of their crops.

Using ground-based photography with a distance of 1.83 m between canopy and camera, greater variation was evident among the relationships between raw digital counts, VIs, and crop response variables for BG canopies. Environmental factors such as time of day, precipitation, and leaf diseases may have affected the strength of these relationships. Standardizing crop response variables has been shown to strengthen correlations between spectral and crop response measurements (Blackmer et al., 1996; Flowers et al., 2001). Additionally, using sun angle as a covariate or normalizing sun angle to 45 degrees could reduce variability associated with taking spectral measurements during the course of a day as sun angle fluctuates (Pinter et al., 1983). Future research should examine these data transformations to determine if they improve the estimation of forage canopy biomass, N concentration, and N uptake using ground-based FCIR photography.

APPENDIX

Appendix 7.1. Treatment means for crabgrass (CG), bermudagrass (BG), volunteer warm season (VWS), and tall fescue (TF) for Kinston 1, 2000.

Forage Species	N Rate	Biomass			N Concentration			N Uptake		
		Mg ha ⁻¹			g kg ⁻¹			kg ha ⁻¹		
		Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
BG (n = 24)	0	1.13	1.73	0.81	12.8	13.4	12.0	14.5	22.0	11.8
	224	3.45	4.80	2.46	14.0	15.1	12.0	48.7	70.5	29.2
	449	4.84	5.63	4.12	14.8	18.1	12.0	72.1	102.0	53.2
	673	5.01	6.04	4.18	18.5	22.3	15.3	93.0	125.1	72.0
CG (n = 21)	0	0.74	1.87	0.34	17.4	24.2	13.6	12.3	29.4	6.6
	224	2.31	4.00	1.51	18.2	24.8	11.2	38.8	48.4	25.3
	449	4.29	5.55	3.42	16.8	22.1	11.9	72.7	111.1	46.6
	673	5.06	5.45	4.81	13.0	15.1	11.2	65.5	72.6	55.2
VWS (n = 19)	0	0.40	0.60	0.22	13.6	17.7	11.8	5.3	8.4	3.0
	224	2.76	5.44	1.01	13.9	17.5	8.9	39.4	93.6	15.5
	449	3.94	4.60	3.30	16.1	23.2	9.7	64.2	97.6	35.4
	673	3.90	4.58	3.33	14.7	16.0	13.9	57.2	63.7	47.7
TF (n = 23)	0	0.25	0.41	0.12	18.4	24.6	10.4	4.4	1.9	8.6
	224	0.85	1.81	0.17	19.2	23.9	10.1	14.3	27.6	3.7
	449	1.55	4.62	0.36	22.1	28.0	13.9	28.8	64.2	7.4
	673	1.71	3.54	0.65	22.7	16.6	32.1	36.0	68.0	12.7

Appendix 7.3. Elemental analysis of swine lagoon effluent fertilizer for Kinston 1 and 2 between January and July of 2001.

Application Dates	Elements										
	N	P	K	Ca	Mg	S	Fe	Mn	Zn	Cu	B
	mg kg ⁻¹										
2/21 to 2/23	452	105	461	279	79	49	23	2.8	8.2	2.7	1.8
3/27	266	71	196	141	53	20	1.6	0.5	0.7	0.2	0.6
5/9 to 5/12	276	92	204	174	60	27	3.9	0.8	1.4	0.4	0.7
6/14 to 6/15	278	96	221	161	61	27	2.6	0.6	1.5	0.2	1.2
Year To Date Average	318	91	270	189	63	30	8	1.2	3.0	0.9	1.1

Appendix 7.4. Means of crop response variables for crabgrass (CG), bermudagrass (BG), volunteer warm season (VWS), and tall fescue (TF) harvested in July, 2000 and fertilized with swine effluent and NH_4NO_3 in Kinston, NC.

N Source		Effluent					NH_4NO_3				
Forage Species	N Rate	Wet Weight	Dry Matter	Biomass	N Conc.	N Uptake	Wet Weight	Dry Matter	Biomass	N Conc.	N Uptake
	kg ha^{-1}	Mg ha^{-1}	g kg^{-1}	Mg ha^{-1}	g kg^{-1}	kg ha^{-1}	Mg ha^{-1}	g kg^{-1}	Mg ha^{-1}	g kg^{-1}	kg ha^{-1}
CG	0	4.53	220	1.00	16	16	2.20	210	0.47	19	9
	224	10.47	210	2.26	16	36	11.40	190	2.37	20	41
	449	17.87	210	3.90	13	51	22.13	210	4.67	20	94
	673	22.66	220	5.06	13	66	‡	‡	‡	‡	‡
BG	0	3.20	430	1.31	13	17	2.66	430	0.95	13	12
	224	7.53	400	2.84	13	37	10.80	390	4.06	15	60
	449	12.13	380	4.38	1.33	58	15.46	360	5.27	16	86
	673	14.47	350	4.85	16	76	15.20	360	5.16	22	110
TF	0	0.93	300	0.27	17	5	0.80	290	0.24	19	4
	224	1.60 [†]	280 [†]	0.46 [†]	20 [†]	8 [†]	4.53	270	1.24	19	21
	449	1.80	280	0.49	22	11	9.40	250	2.26	22	41
	673	4.80	270	1.25	11	24	9.32	230	2.16	25	48
VWS	0	1.73	250	0.44	13	5	1.46	230	0.36	15	5
	224	5.20	260	1.36	13	17	23.00 [†]	210 [†]	4.87 [†]	15 [†]	73 [†]
	449	15.13	260	3.85	12	44	19.10 [†]	210 [†]	4.07 [†]	23 [†]	94 [†]
	673	17.73	220	3.90	15	57	‡	‡	‡	‡	‡

[†]Consists of only 2 replications

‡ Eliminated due to tissue injury

Appendix 7.5. Treatment means for raw digital counts and vegetation indices for bermudagrass canopies harvested at early heading in September, 2000 in Raleigh, NC.

N Rate kg ha ⁻¹	Spectral Variables									
	G	R	NIR	NDVI	Norm NIR	NR	NG	RVI	DVI	GNDVI
0	124	114	129	0.066	0.351	0.308	0.341	1.143	16	0.015
11	120	107	132	0.113	0.369	0.294	0.337	1.261	25	0.045
22	113	103	129	0.106	0.371	0.299	0.330	1.245	26	0.057
45	127	115	150	0.137	0.384	0.292	0.325	1.320	35	0.084
90	137	129	162	0.118	0.380	0.299	0.321	1.272	33	0.083

Appendix 7.6. Treatment means for raw digital counts and vegetation indices for bermudagrass canopies harvested and photographed in July, 2000 and 2001 at Site 2S in Sampson Co., NC.

RYE	Variable	N	Mean	Std. Dev.	Min	Max
0	G	8	136	29	93	180
	R	8	116	32	68	174
	NIR	8	135	27	106	184
	NDVI	8	0.0848	0.0825	-0.0641	0.2076
	NormNIR	8	0.3522	0.0312	0.2913	0.3943
	RVI	8	1.2	0.19	0.88	1.52
	DVI	8	19	18	-16	40
	NR	8	0.2965	0.0233	0.2587	0.3312
	NG	8	0.3512	0.0136	0.3342	0.3775
	GNDVI	8	0.0001	0.0613	-0.129	0.0638
75	G	8	130	31	93	171
	R	8	95	33	60	143
	NIR	8	150	26	119	197
	NDVI	8	0.2416	0.1077	0.0887	0.3438
	NormNIR	8	0.4068	0.0403	0.3464	0.4453
	RVI	8	1.68	0.36	1.19	2.05
	DVI	8	55	20	26	82
	NR	8	0.2479	0.0322	0.2175	0.2946
	NG	8	0.3452	0.0124	0.3342	0.3724
	GNDVI	8	0.0797	0.0651	-0.0361	0.1382
100	G	8	130	43	80	179
	R	8	99	47	48	155
	NIR	8	156	33	118	204
	NDVI	8	0.2317	0.1473	0.0924	0.4479
	NormNIR	8	0.4198	0.0544	0.3592	0.4908
	RVI	8	1.8105	0.5858	1.2035	2.6224
	DVI	8	57	17	31	77
	NR	8	0.248	0.0462	0.1872	0.2993
	NG	8	0.3353	0.0102	0.322	0.3508
	GNDVI	8	0.1078	0.0763	0.0448	0.2077
125	G	8	131	27	92	162
	R	8	92	29	55	129
	NIR	8	160	24	134	194
	NDVI	8	0.2848	0.0918	0.1457	0.418
	NormNIR	8	0.4219	0.0337	0.3747	0.4769
	RVI	8	1.838	0.372	1.341	2.436
	DVI	8	68	12	44	79
	NR	8	0.2345	0.0292	0.1957	0.2794
	NG	8	0.3434	0.0121	0.3274	0.3691
	GNDVI	8	0.1011	0.0505	0.0357	0.1858

RYE	Variable	N	Mean	Std. Dev.	Min	Max
200	G	8	143	20	96	162
	R	8	102	19	61	130
	NIR	8	166	17	130	180
	NDVI	8	0.2441	0.0668	0.1384	0.3624
	NormNIR	8	0.4065	0.0263	0.3698	0.4536
	RVI	8	1.664	0.243	1.321	2.136
	DVI	8	64	13	42	78
	NR	8	0.2467	0.0194	0.2123	0.2799
	NG	8	0.3467	0.0084	0.3341	0.3618
	GNDVI	8	0.0784	0.0428	0.0221	0.1517

Appendix 7.7. Treatment means for raw digital counts and vegetation indices for bermudagrass canopies harvested and photographed in July and August, 2000 at Site 2S in Sampson Co., NC.

RYE	Variable	N	Mean	Std. Dev.	Min	Max
0	G	8	131	23	93	161
	R	8	120	34	69	167
	NIR	8	139	24	106	172
	NDVI	8	0.0841	0.0648	0.0138	0.2076
	NormNIR	8	0.3590	0.0171	0.3433	0.3943
	RVI	8	1.19	0.16	1.03	1.52
	DVI	8	19	11	5	36
	NR	8	0.3037	0.026	0.2587	0.334
	NG	8	0.3373	0.0131	0.3216	0.3507
	GNDVI	8	0.0309	0.0224	-0.0059	0.0638
75	G	8	122	24	93	150
	R	8	101	40	60	146
	NIR	8	147	21	119	174
	NDVI	8	0.2147	0.1287	0.0755	0.3438
	NormNIR	8	0.4055	0.0373	0.3655	0.4453
	RVI	8	1.61	0.42	1.163	2.047
	DVI	8	47	21	24	70
	NR	8	0.2631	0.0470	0.2175	0.3150
	NG	8	0.3313	0.0104	0.3186	0.3419
	GNDVI	8	0.0991	0.0323	0.0609	0.1382
100	G	8	117	31	80	155
	R	8	98	45	48	149
	NIR	8	148	25	118	183
	NDVI	8	0.2442	0.1626	0.0750	0.4479
	NormNIR	8	0.4199	0.0533	0.3661	0.4908
	RVI	8	1.76	0.63	1.16	2.62
	DVI	8	50	22	22	77
	NR	8	0.2554	0.0564	0.1872	0.3150
	NG	8	0.3245	0.0086	0.3186	0.3445
	GNDVI	8	0.1244	0.0586	0.0689	0.2077
125	G	8	120	15	92	138
	R	8	93	27	55	129
	NIR	8	150	10	134	166
	NDVI	8	0.2512	0.1216	0.1198	0.4180
	NormNIR	8	0.4183	0.0364	0.3785	0.4769
	RVI	8	1.73	0.46	1.27	2.43
	DVI	8	57	21	33	79
	NR	8	0.2510	0.0437	0.1957	0.2982
	NG	8	0.3306	0.0114	0.3182	0.3479
	GNDVI	8	0.1156	0.0354	0.0776	0.1858

RYE	Variable	N	Mean	Std. Dev.	Min	Max
200	G	8	135	21	96	156
	R	8	105	20	61	124
	NIR	8	159	18	130	179
	NDVI	8	0.2087	0.0881	0.1059	0.3624
	NormNIR	8	0.4005	0.0268	0.3726	0.4536
	RVI	8	1.55	0.31	1.23	2.14
	DVI	8	54	18	27	70
	NR	8	0.2625	0.0319	0.2123	0.3024
	NG	8	0.3368	0.0124	0.3197	0.3513
	GNDVI	8	0.0857	0.0330	0.0545	0.1517

Appendix 7.8. Treatment means for raw digital counts and vegetation indices for bermudagrass canopies harvested and photographed at four locations in August, 2000 in NC.

RYE	Variable	N	Mean	Std. Dev.	Min	Max
0	G	8	135	16	103	161
	R	8	131	21	90	167
	NIR	8	149	21	115	190
	NDVI	8	0.0656	0.0565	0.0026	0.1661
	NormNIR	8	0.3594	0.0213	0.3359	0.4028
	RVI	8	1.14	0.13	1.00	1.39
	DVI	8	18	16	0.66	49
	NR	8	0.3149	0.0176	0.2866	0.3343
	NG	8	0.3255	0.0070	0.3091	0.3372
	GNDVI	8	0.0486	0.0370	0.0086	0.1316
75	G	8	133	14	100	150
	R	8	124	16	86	146
	NIR	8	160	17	119	186
	NDVI	8	0.1298	0.0380	0.0755	0.2057
	NormNIR	8	0.3852	0.0139	0.3655	0.4096
	RVI	8	1.30	0.10	1.16	1.52
	DVI	8	36	10	24	53
	NR	8	0.2966	0.0125	0.2699	0.3150
	NG	8	0.3180	0.0050	0.3088	0.3271
	GNDVI	8	0.0952	0.0225	0.0609	0.1309
100	G	8	121	24	69	155
	R	8	111	26	57	149
	NIR	8	149	28	75	183
	NDVI	8	0.1491	0.0454	0.0750	0.2200
	NormNIR	8	0.3911	0.0172	0.3661	0.4192
	RVI	8	1.35	0.12	1.16	1.56
	DVI	8	38	10	18	53
	NR	8	0.2895	0.0150	0.2654	0.3150
	NG	8	0.3193	0.0081	0.3094	0.3439
	GNDVI	8	0.1006	0.0301	0.0417	0.1455
125	G	8	126	10	103	138
	R	8	116	13	92	134
	NIR	8	156	14	131	182
	NDVI	8	0.1498	0.0384	0.0809	0.2162
	NormNIR	8	0.3929	0.0141	0.3689	0.4172
	RVI	8	1.35	0.11	1.17	1.55
	DVI	8	40	10	20	58
	NR	8	0.2905	0.0131	0.2681	0.3137
	NG	8	0.3164	0.0064	0.3084	0.3318
	GNDVI	8	0.1075	0.0235	0.0682	0.1499

RYE	Variable	N	Mean	Std. Dev.	Min	Max
200	G	8	123	13	100	150
	R	8	112	15	82	141
	NIR	8	153	16	131	188
	NDVI	8	0.1593	0.0395	0.1033	0.2236
	NormNIR	8	0.3953	0.0153	0.3726	0.4151
	RVI	8	1.38	0.11	1.23	1.57
	DVI	8	42	9	27	57
	NR	8	0.2866	0.0133	0.2629	0.3088
	NG	8	0.3180	0.0092	0.3015	0.3341
	GNDVI	8	0.1080	0.0292	0.0545	0.1585