Remote Sensing of Environment

Estimating Corn Leaf Chlorophyll Concentration from Leaf and Canopy Reflectance

C. S. T. Daughtry, C. L. Walthall,* M. S. Kim,† E. Brown de Colstoun‡ and J. E. McMurtrey III**

*Farmers must balance the competing goals of supplying adequate N for their crops while minimizing N losses to the environment. To characterize the spatial variability of N over large fields, traditional methods (soil testing, plant tissue analysis, and chlorophyll meters) require many point samples. Because of the close link between leaf chlorophyll and leaf N concentration, remote sensing techniques have the potential to evaluate the N variability over large fields quickly. Our objectives were to (1) select wavelengths sensitive to leaf chlorophyll concentration, (2) simulate canopy reflectance using a radiative transfer model, and (3) propose a strategy for detecting leaf chlorophyll status of plants using remotely sensed data. A wide range of leaf chlorophyll levels was established in field-grown corn (*Zea mays *L.) with the application of 8 N levels: 0%, 12.5%, 25%, 50%, 75%, 100%, 125%, and 150% of the recommended rate. Reflectance and transmittance spectra of fully expanded upper leaves were acquired over the 400-nm to 1,000-nm wavelength range shortly after anthesis with a spectroradiometer and integrating sphere. Broad-band differences in leaf spectra were observed near 550 nm, 715 nm, and >750 nm. Crop canopy reflectance was simulated using the SAIL (Scattering by Arbitrarily Inclined Leaves) canopy reflectance model for a wide range of background reflectances, leaf area indices (LAI), and leaf chlorophyll concentrations. Variations in background reflectance and LAI confounded the detection of the relatively subtle differences in canopy reflectance due to changes in leaf chlorophyll concentration. Spectral vegetation indices that combined near-infrared reflectance and red reflectance (e.g., OSAVI and NIR/Red) minimized contributions of background reflectance, while spectral vegetation indices that combined reflectances of near-infrared and other visible bands (MCARI and NIR/Green) were responsive to both leaf chlorophyll concentrations and background reflectance. Pairs of these spectral vegetation indices plotted together produced isolines of leaf chlorophyll concentrations. The slopes of these isolines were linearly related to leaf chlorophyll concentration. A limited test with measured canopy reflectance and leaf chlorophyll data confirmed these results. The characterization of leaf chlorophyll concentrations at the field scale without the confounding problem of background reflectance and LAI variability holds promise as a valuable aid for decision making in managing N applications.* *Published by Elsevier Science Inc.*

INTRODUCTION

Nitrogen is an essential element for plant growth and is frequently the major limiting nutrient in most agricultural soils. Profitable corn (*Zea mays* L.) production systems require inputs of large quantities of N. Nitrogen fertilizer in excess of a crop's nutritional needs may move into surface water and groundwater and contribute to eutrophication of lakes and streams (Wood et al., 1993). Farmers must balance the competing goals of supplying enough N to their crops while minimizing the loss of N to the environment, which represents both a threat to water quality and an economic loss. The economic penalties of reduced yields from supplying inadequate N are substantial.

Traditionally, soil testing, plant tissue analysis, and long-term field trials have been used for assessing N availability for crops. More recently, chlorophyll meters (e.g., SPAD-502,[1] Minolta Osaka Co., Ltd., Japan) have been

*USDA, ARS, Remote Sensing and Modeling Laboratory
†USDA, ARS, Instrumentation and Sensing Laboratory
‡Raytheon STX, Lanham, Maryland

Address correspondence to C. S. T. Daughtry, Remote Sensing and Modelling Lab., USDA/ARS, 10300 Baltimore Avenue, Beltsville, MD 20705-2350, USA. E-mail: cdaughtry@asrr.arsusda.gov
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[1] Company and product names are used for clarity and do not imply any endorsement by USDA to the exclusion of other comparable products.

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used to monitor plant N status by measuring transmittance of radiation through a leaf in two wavelength bands centered near 650 nm and 940 nm (e.g., Peterson et al., 1993; Blackmer et al., 1994; Wood et al., 1993). Leaf chlorophyll meter readings, which are essentially a measure of leaf greenness, are generally linear with extractable chlorophyll concentrations for a wide variety of crops. However, plant species and/or environmental conditions may affect the relationship (Wood et al., 1993). Markwell et al. (1995) theoretically justified the use of an exponential equation that forces a more appropriate fit to limited data sets than polynomial equations.

Because the majority of leaf N is contained in chlorophyll molecules, there is a close link between leaf chlorophyll content and leaf N content (Yoder and Pettigrew-Crosby, 1995). However, at high N levels, the relationship between leaf chlorophyll and leaf N concentration may be nonlinear, indicating the presence of nonchlorophyll N, probably NO_3-N (Wood et al., 1993).

Although the chlorophyll meter allows a relative N assessment without time-consuming laboratory analysis, it required taking 30 or more readings from representative plants in each area of interest (Peterson et al., 1993), which can be time-consuming for large fields that have spatial variations in N. Blackmer and Schepers (1995) recommended interpreting the variability in chlorophyll meter readings relative to reference areas in each field, where N is not limiting. These in-field reference areas helped to normalize differences among cultivars, growth stages, and environmental conditions.

Remote sensing techniques, based on measuring the reflected radiation from plant canopies, have the potential of evaluating the N status of many plants within the field of view of the sensor. Spectral reflectance measurements of corn and wheat (*Triticum aestivium* L.) canopies have been used to detect differences in N treatments (Blackmer et al., 1996; Walburg et al., 1982; Hinzman et al., 1986). Differences in leaf area index, biomass, foliage cover, and leaf chlorophyll content resulted from the N treatments and contributed to the differences in canopy spectral reflectance observed. The relatively subtle differences in canopy reflectance associated with changes in leaf chlorophyll are often confounded with major changes in plant growth and development due to N treatments.

Blackmer et al. (1996) calculated the reflectance of corn canopies relative to reference canopies supplied with nonlimiting available N. They found that the ratio of reflected radiation in a visible band (550–600 nm) and a near-infrared band (800–900 nm) could be used to detect N stress in corn. The use of relative reflectance helped reduce variability in the measurements. They speculated that the characterization of the spatial patterns in canopy reflectance can be the basis for developing technologies for the application of N at variable rates to agricultural fields.

Our objectives were (1) to select wavelengths sensitive to leaf chlorophyll concentration; (2) to simulate canopy reflectance, using a radiative transfer model, for a range of leaf optical properties, leaf area indices, and soil backgrounds; and (3) to propose a strategy for detecting N status of crops using remotely sensed data.

MATERIALS AND METHODS

Corn (*Zea mays* L.) was grown on a Woodstown sandy loam soil devoted to long-term N application rate studies at Beltsville, Maryland. The field was maintained at high soil test levels of P and K. Ammonium nitrate was applied after planting to supply 0%, 12.5%, 25%, 50%, 75%, 100%, 125%, and 150% of the recommended rate of 180 kg N/ha and to establish a range of leaf chlorophyll levels. Plot size was 6 m×15 m and each N treatment was replicated twice. Grain from each plot was hand-harvested at maturity, shelled, dried, and weighed. McMurtrey et al. (1994) described the experiment in detail.

Leaf Spectra

Shortly after anthesis (stage R1), fully expanded leaves near the tops of 10 plants in each plot were excised, placed in a plastic bag in an ice chest, and transported to the laboratory for spectral measurements. Leaf reflectance and transmittance were measured with an integrating sphere (Model LI-1800, LiCor, Inc., Lincoln, NE, USA) coupled to a spectroradiometer (Model SE590, Spectron Engineering, Inc., Denver, CO, USA) over the 400-nm to 1,000-nm wavelength range at approximately 3-nm intervals. Both adaxial and abaxial surfaces of each leaf were measured and reflectance and transmittance factors were calculated (Daughtry et al., 1989). After the spectral measurements, a 131-mm^2 disk was cut from each leaf for pigment analysis. Each leaf disk was extracted for 24 hours in the dark at 25°C with dimethyl sulfoxide (DMSO) after which absorptance was measured and concentrations of chlorophyll *a* and chlorophyll *b* were computed using the equations of Lichtenthaler (1987).

Soil and Crop Residue Spectra

Topsoil samples from three U.S. cropland soils, selected to span the wide range of reflectance expected in most agricultural fields, are listed in Table 1. The soils were dried, crushed, and passed through a 2-mm sieve. Corn and soybean (*Glycine max* Merr.) residues were collected 8 months after harvest from fields near Beltsville, Maryland. Dried soil and crop residues were placed in 45-cm square sample trays. Reflectance spectra were acquired outdoors near solar noon under clear sky conditions using a spectroradiometer (Model SE590, Spectron Engineering, Denver, CO, USA) with a 1° field of view. The spectroradiometer was mounted with a nadir view at 0.7 m above the sample trays. Twenty locations per sample tray were measured. After the spectra of the dry samples were acquired, the samples were thoroughly wetted with water and allowed to drain, and a second set of spectra was acquired. The spectra were referenced to a Spectralon panel (Labsphere,

Table 1. Input Data for the SAIL Model

Input Parameters	*Input Values*
Leaf reflectance and transmittance	Low, medium, and high chlorophyll concentrations (18.3, 35.0, and 52.4 $\mu g/cm^2$, respectively)
Background reflectance (dry and wet)	Barnes (coarse-loamy, mixed Udic Haploboroll), from Morris, Minnesota
	Othello (fine-silty, mixed, mesic Typic Ochraquult), from Salisbury, Maryland
	Cecil (clayey, kaolinitic, thermic, Typic Hapludult), from Watkinsville, Georgia
	corn residue, 8 months after harvest, from Beltsville, Maryland
	soybean residue, 8 months after harvest, from Beltsville, Maryland
LAI	0, 0.01, 0.1, 0.5, 1, 1.5, 2, 3, 4, 6, 8
Leaf angle distribution	Spherical
View zenith angle	0 degrees (nadir)
Sun zenith angle	45 degrees
Fraction of direct incoming radiation	1.0

Inc., North Sutton, NH, USA) and reflectance factors were calculated (Walter-Shea and Biehl, 1990).

Simulated Canopy Reflectance

Canopy reflectance was simulated using the SAIL (Scattering by Arbitrarily Inclined Leaves) model (Verhoef, 1984). The SAIL model is a turbid-medium model that treats the canopy as a horizontally uniform plane-parallel layer with diffusely reflecting and transmitting elements (Goel, 1989). Canopy architecture is expressed through leaf area index (LAI) and leaf angle distribution. The soil or lowest layer is assumed to be a diffuse reflector. Input spectral data included mean reflectance and transmittance of leaves with low, medium, and high chlorophyll concentrations and reflectance of three soils and two crop residues at two moisture conditions (Table 1). Although the fraction of direct radiation was set rather high at 1.0, it had little effect on the comparisons. Other input conditions are listed in Table 1. Canopy reflectance factors were simulated for LAIs of 0, 0.01, 0.1, 0.5, 1, 1.5, 2, 3, 4, 6, and 8. Difference spectra were calculated by subtracting the canopy reflectance spectra of the high (100%) N treatment from the canopy reflectance spectra of selected other treatments. Spectral vegetation indices were computed using narrow spectral bands for each background (five soils×two moistures), chlorophyll concentrations (three levels), and LAI (11 values). Several spectral vegetation indices were computed using narrow-band reflectance factors and were normalized between 0 for the minimum value and 1 for the maximum value. Performance of the spectral vegetation indices were evaluated using analysis of variance (Daughtry et al., 1980).

Measured Canopy Reflectance

Canopy reflectance data from Walburg et al. (1982) were used to verify the results from the SAIL model. Briefly, corn was grown on a Raub silt loam (Aquic Argiudoll) during 1979 at the Purdue Agronomy Farm near West Lafayette, Indiana. The long-term N treatments were 0 kg N/ha, 67 kg N/ha, 134 kg N/ha, and 202 kg N/ha applied in the spring. Spectral reflectance over the 400-nm to 2,400-nm wavelength range were acquired with a spectroradiometer (Model 20C, Exotech, Inc., Gaithersburg, MD, USA) positioned 9.1 m above the plots with a 15° field of view. Multispectral data were acquired near solar noon on 12 dates during the growing season under clear skies and referenced to painted $BaSO_4$ panel. Agronomic variables measured included LAI, biomass, percent cover, and leaf chlorophyll concentration. Walburg et al. reported the spectral data as band means corresponding to Landsat Thematic Mapper (TM) bands: TM2 (520–600 nm), TM3 (630–690 nm), and TM4 (760–900 nm). Spectral vegetation indices identified during analysis of the SAIL data were computed using the Landsat TM bands.

RESULTS AND DISCUSSION

Leaf Reflectance and Transmittance

Mean leaf chlorophyll concentrations are presented in Table 2. Total chlorophyll concentrations more than doubled

Table 2. Mean Leaf Chlorophyll Concentration and Grain Yields of Corn Fertilized with Different Rates of N

N Applied		*Chlorophyll a*	*Chlorophyll b*	*Total Chlorophyll*	*Grain Yield*	
kg/ha	*%*	*($\mu g/cm^2$)*	*($\mu g/cm^2$)*	*($\mu g/cm^2$)*	*(kg/ha)*	*Relative Yield*
0	0	10.4	7.9	18.3	1478	0.38
22	12	14.8	7.3	22.1	1651	0.43
45	25	14.6	6.9	21.5	2000	0.52
90	50	24.2	10.8	35.0	3759	0.97
135	75	34.8	16.1	50.9	3682	0.95
180	100	36.3	16.1	52.4	3877	1.00
225	125	35.1	16.1	51.3	4615	1.19
270	150	34.6	15.0	49.5	3750	0.97

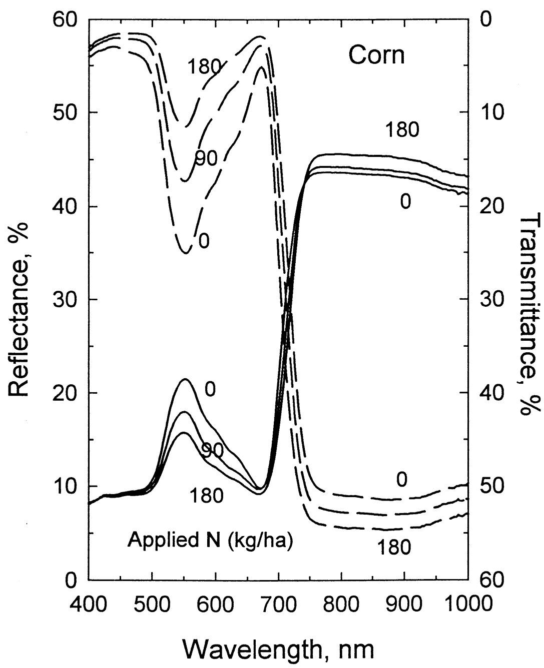


Figure 1. Leaf reflectance and transmittance for 0 180 kgN/ha, 90 180 kgN/ha, and 180 kgN/ha (i.e., 0%, 50%, and 100% of recommended rate).

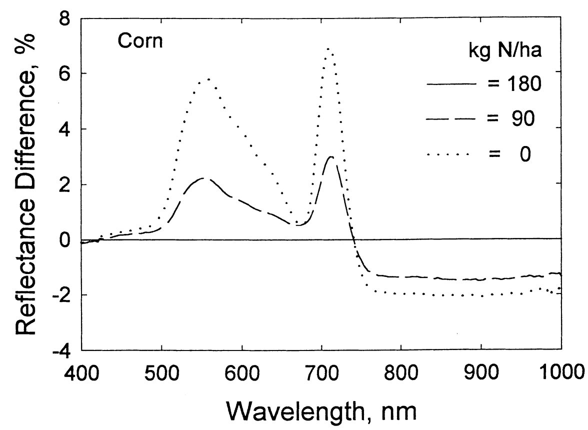


Figure 2. Leaf reflectance and transmittance difference spectra relative to 180 kgN/ha (100% N) rate.

as N fertilizer application rates increased. Chlorophyll concentrations were not significantly different at N rates above 135 kg/ha or 75% of the recommended management practice. Grain yields increased rapidly as more N fertilizer was applied but plateaued at 180 kg N/ha. Water was also limited during late grain filling and probably contributed to the lack of differences in grain yields (McMurtrey et al., 1994). Leaf spectra were acquired 2 weeks before water stress symptoms were visible.

Mean reflectance and transmittance of 10 leaves from plants fertilized with three rates of N are shown in Fig. 1. The other rates were intermediate and were not shown for clarity. Leaves with the lowest chlorophyll contents had the highest reflectance and transmittance in the visible (400–700 nm) and lowest reflectance and transmittance in the near-infrared (700–1000 nm). Leaf reflectance at 550 nm ranged from 15.8% for leaves with high chlorophyll levels to 21.5% for leaves with low chlorophyll levels (Fig. 1).

In the blue (450 nm) and red (670 nm) wavelength regions, light absorption by chlorophyll clearly dominated the spectral properties of the leaves (Chappelle et al., 1992). Leaf transmittance showed greater changes with N fertilization (and chlorophyll concentration) than leaf reflectance. All of the transmitted light must pass through the leaf, increasing the likelihood of its interaction with chlorophyll and other light-absorbing molecules. However, a portion of the radiation striking a leaf is reflected at the first leaf surface and never interacts with leaf pigments and internal leaf structures (Grant, 1987). This first surface reflectance effectively adds an offset to the reflected signal that is not present in transmittance. For this reason, chlorophyll meters typically measure leaf transmittance, which is inversely related to chlorophyll concentration (Markwell et al., 1995).

To highlight where leaf chlorophyll content affected leaf optical properties, the reflectance and transmittance spectra were normalized by the mean spectrum of the well-fertilized leaves (180 Kg N/ha). Broad-band differences are evident near 550 nm, 715 nm, and >750 nm in reflectance (Fig. 2). Differences in the blue (450 nm) and red (670 nm) were small (<1%) even though chlorophyll concentration nearly tripled (Table 2). Apparently, chlorophyll concentrations were sufficient to absorb nearly all of the blue and red radiation. Reflectance in the green (550 nm) and red-edge (715 nm) bands increased significantly as chlorophyll concentration decreased (Fig. 2).

Reflectance at selected wavelengths is plotted as a function of total leaf chlorophyll concentration (Fig. 3). Reflectance factors at 550 nm and 715 nm were inversely related to chlorophyll concentrations, while reflectance factors at 450 nm and 670 nm changed only slightly. Reflectance factors in the near-infrared (>750 nm) were not related to leaf chlorophyll, but to leaf structure (Knipling, 1970). Similar relationships have been reported for numerous woody and herbaceous species (Chappelle et al., 1992; Gitelson and Merzlyak, 1997).

Background Reflectance

Reflectance spectra of the wet and dry soil and crop residues, used for simulating the reflectance of the corn canopies, are shown in Fig. 4. When moisture content increased, reflectance decreased at all wavelengths. The changes in reflectance were associated with changes that occur when the air-particle interfaces were replaced by air-water-particle interfaces (Irons et al., 1992). These backgrounds spanned a wide range of reflectance factors at all wavelengths. For example, in the green region at 550 nm, soil

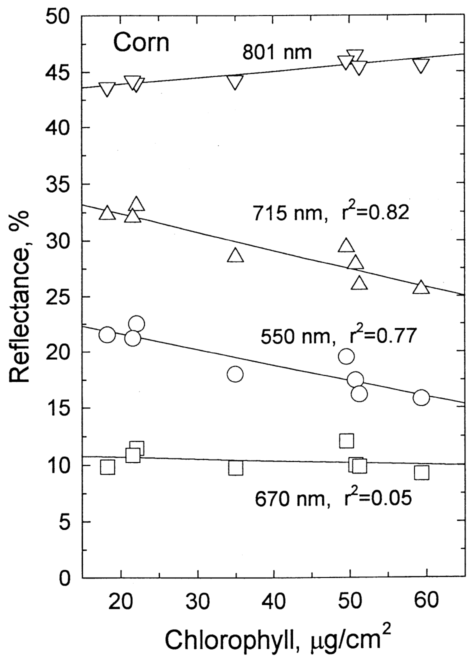


Figure 3. Relationship of leaf chlorophyll concentration to reflectance at selected wavelengths.

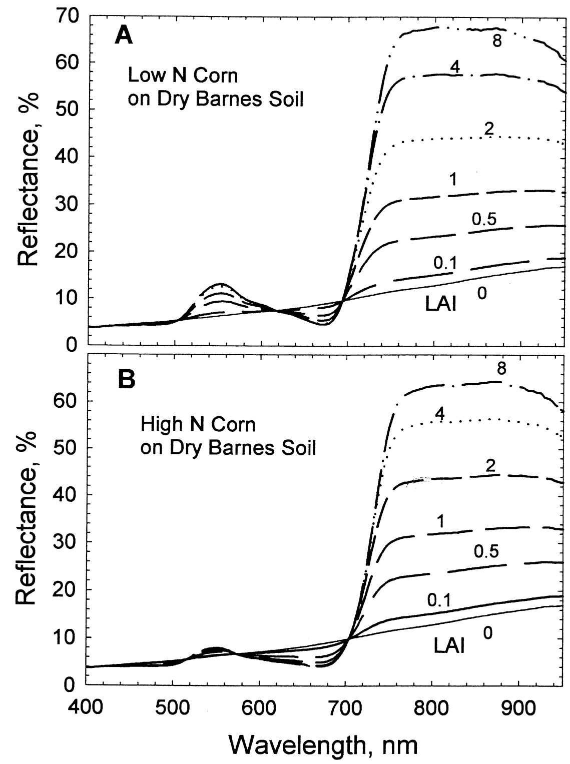
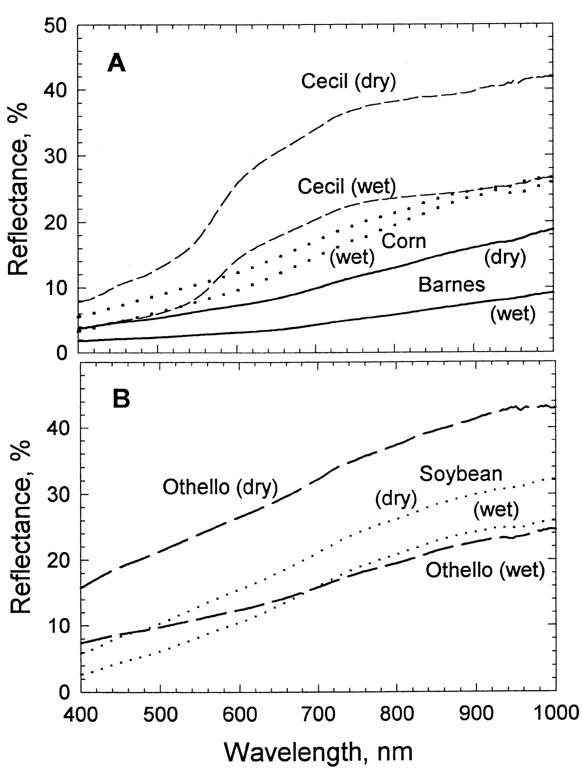


Figure 5. Simulated scene reflectance spectra of (A) low N and (B) high N corn on dry Barnes soil. Reflectance spectra are shown for LAI=0 (bare soil), 0.1, 0.5, 1, 2, 4, and 8.

Figure 4. Reflectance of Barnes, Cecil, and Othello soils plus corn and soybean crop residues. For each soil and crop residue, the upper line is the dry reflectance spectrum and the lower line is the wet reflectance spectrum.



reflectance ranged from 2.7% for wet Barnes to 23.6% for dry Othello, a nearly nine-fold change in reflectance (Fig. 4).

Simulated Canopy Reflectance

While leaf reflectance is important for assessing individual plants, canopy reflectance is required for assessing the spatial variability of crop conditions in fields. Many factors complicate the transition from leaf reflectance to canopy reflectance. As leaf area per unit ground area or LAI increases, the contribution of the background (i.e., soil or crop residue) to the overall scene reflectance decreases and the total amount of radiation scattered by plant cells increases (Norman et al., 1985). Multiple scattering is particularly important in the near-infrared (700–1300 nm) where plant pigments do not absorb radiation (Knipling, 1970; Chappelle et al., 1992; Gitelson et al., 1996).

Reflectance spectra of 330 corn canopies were simulated with the SAIL model (Verhoef, 1984) using the input data in Table 1. With the SAIL model, we changed to the soil underneath the corn canopies and estimated the contribution of soil reflectance to canopy reflectance. Figs. 5 and 6 show reflectance spectra of corn canopies composed of leaves with either a low or a high chlorophyll concentration for a range of LAI values. The dry, bare Barnes soil was darker than the simulated corn canopies at 550 nm and in the near-infrared at >720 nm (Fig. 5). The

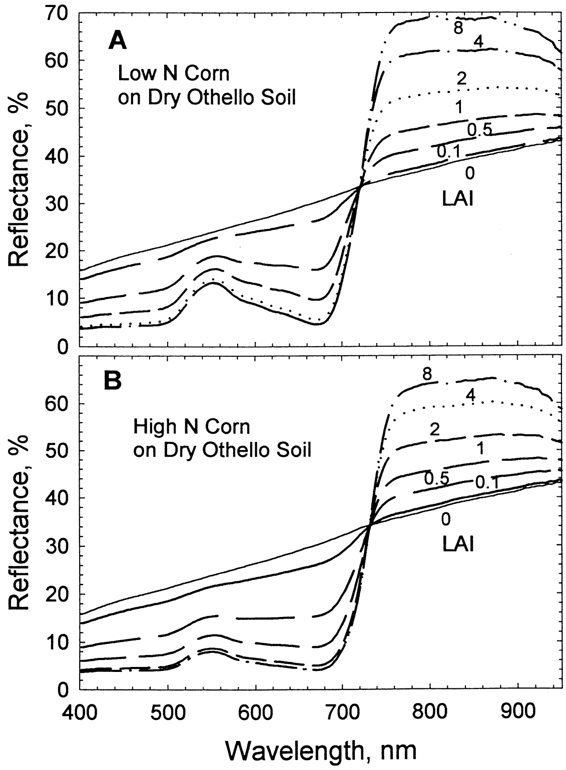


Figure 6. Simulated scene reflectance spectra of (A) low N and (B) high N corn on dry Othello soil. Reflectance spectra are shown for LAI=0 (bare soil), 0.1, 0.5, 1, 2, 4, and 8.

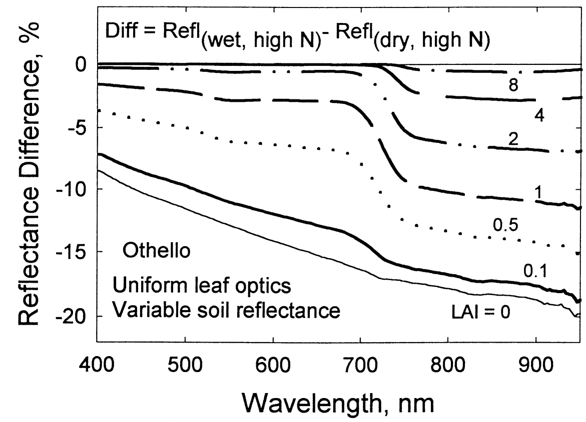


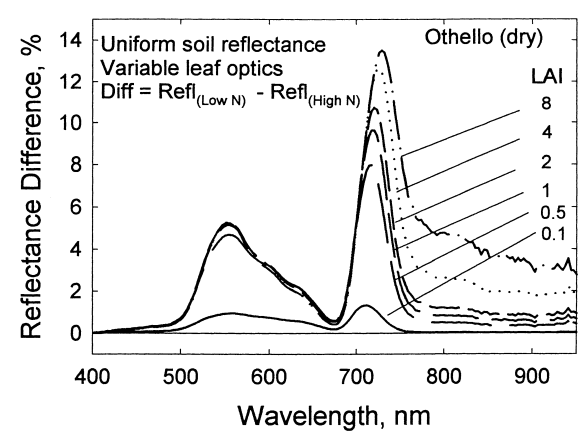
Figure 7. Scene reflectance difference spectra for variable soil reflectance(wet Othello vs. dry Othello) and uniform leaf optics (high leaf N). Difference=Reflectance(High N, wet soil)−Reflectance(High N, dry soil). The horizontal line at 0 indicates no differences in reflectance due to soil moisture changes.

addition of leaves on the dry Barnes soil slightly increased canopy reflectance in the visible wavelengths, but rapidly increased canopy reflectance in the near-infrared. The dry, bare Othello soil was brighter than the corn canopies in the visible (400–720 nm), but was slightly darker than the corn canopies in the near-infrared (Fig. 6). The other backgrounds were intermediate and were not shown for brevity. The reflectance spectra, presented in Figs. 5 and 6, illustrate how profoundly background reflectance can affect canopy reflectance, particularly for LAIs less than 2. Thus, attempts to assess plant nutrient status based on canopy reflectance in a single band often will be confounded by the variability in background reflectance and/or LAI.

Although Barnes and Othello soils are unlikely to ever occur together in the same field, significant variability in reflectance due to differences in soil moisture across a field is likely to occur. Fig. 7 illustrates how soil brightness changes due to surface soil moisture affect canopy reflectance for a range of leaf area indices. For each LAI in Fig. 7, the difference spectra were normalized by the reflectance spectrum of the canopy with high chlorophyll content on the dry soil. The horizontal line at 0 indicates no differences in reflectance due to soil moisture. At LAI <2, the differences in canopy reflectance due solely to soil moisture (soil color) conditions exceeded 1% (absolute reflectance units) across all wavelengths and represent a major source of variation in canopy reflectance, particularly in the visible wavelengths where reflectance is generally less than 30% (Fig. 6). Surface soil moisture changes across a field due to progressive irrigation or uneven soil drying introduce significant variations in canopy reflectance, even for uniform crop canopies, and confound detection and identification of plant stresses.

In Fig. 8, canopy reflectance differences due solely to variable leaf reflectance and transmittance (associated with changes in leaf chlorophyll concentration) are shown for a constant soil reflectance (dry Othello). Major differences in canopy reflectance due solely to changes in leaf optics occurred in two broad bands near 550 nm and 715 nm. Reductions in leaf chlorophyll concentration increased can-

Figure 8. Scene reflectance difference spectra for uniform soil reflectance (dry Othello) and variable leaf optics (low leaf N vs. high leaf N). Difference=Reflectance(Medium N, dry soil)−Reflectance(High N, dry soil). The horizontal line at 0 indicates no differences in reflectance due to changes in leaf chlorophyll concentrations.



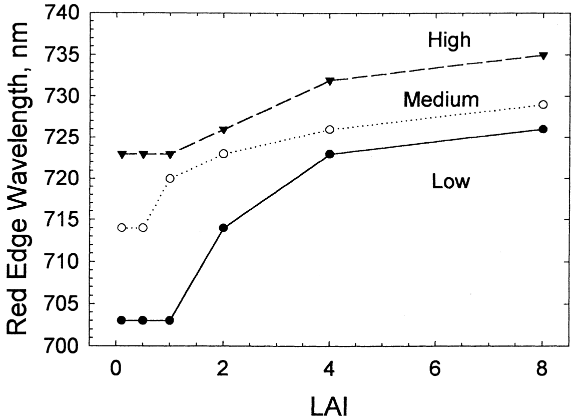


Figure 9. Change in the red-edge position as a function of LAIs and leaf chlorophyll concentration.

opy reflectance at 550 nm. The differences in canopy reflectance near 715 nm were associated with the transition from chlorophyll absorption processes in the red wavelengths to within-leaf scattering in the near-infrared wavelengths (Munden et al., 1994). In Fig. 8, the maximum difference near 715 nm shifted to longer wavelengths as LAI and chlorophyll density increased. The position of this transition is often referred to as the "red-edge" and is defined as the wavelength of the maximum slope (maximum first derivative) of the reflectance spectrum in the 650-nm to 800-nm wavelength region (Horler et al., 1983). An increase in chlorophyll concentration caused a broadening of the chlorophyll absorption feature in the red and consequently moved the position of the red-edge to longer wavelengths (Munden et al., 1994). For these simulated corn canopies, the position of the red-edge shifted toward longer wavelengths as both leaf chlorophyll concentration increased and LAI increased for all backgrounds (Fig. 9).

From Figs. 5 to 9, it is clearly evident that LAI, background reflectance, and leaf chlorophyll concentration interact and modulate canopy reflectance. To assess leaf chlorophyll (and leaf N) status from remotely sensed observations, spectral indices are needed that are sensitive to leaf chlorophyll concentration and minimize variations in canopy reflectance associated with background reflectance and LAI. Blackmer et al. (1996) recommended using in-field reference areas with nonlimiting available N to normalize for differences in backgrounds and growth stages. However, this approach requires applying excess N to portions of each field and assuming that the background reflectance and crop growth are identical between reference and nonreference areas of the fields.

Spectral Vegetation Indices

Spectral vegetation indices use the characteristic shape of the green vegetation spectrum by combining the low reflectance in the visible wavelengths with the high reflectance of the near-infrared wavelengths. The combinations may be ratios of two or more bands, slopes, or other formulations that minimize variation due to extraneous factors and maximize sensitivity to the variable of interest. The reported vegetation indices are numerous and may be broadly grouped into three categories: (1) intrinsic indices, (2) soil-line related indices, and (3) atmospherically adjusted indices (Rondeaux et al., 1996; Baret and Guyot, 1991). In this paper, we will focus on examples of intrinsic and soil-line vegetation indices only.

Intrinsic indices rely solely on the measured spectral reflectance data and include ratios of two or more bands in the visible and near-infrared wavelengths. The simple ratio is the most common [see Eq. (1)].

$$\mathrm{NIR/Red} = R_{801}/R_{670} \tag{1}$$

where R_{801} is the near-infrared band reflectance factor at 801 nm and R_{670} is the red-band reflectance factor at 670 nm. The actual wavelength bands used in the indices vary from sensor to sensor but include a visible band and a near-infrared band. Another simple ratio is NIR/Green [see Eq. (2)]

$$\mathrm{NIR/Green} = R_{801}/R_{550} \tag{2}$$

where R_{550} is the green-band reflectance factor at 550 nm. The intrinsic group also includes the Normalized Difference Vegetation Index (NDVI), as seen in Eq. (3):

$$\mathrm{NDVI} = (R_{801} - R_{670})/(R_{801} + R_{670}) \tag{3}$$

and the Green Normalized Difference Vegetation Index (Green NDVI), as seen in Eq. (4)

$$\mathrm{Green\ NDVI} = (R_{801} - R_{550})/(R_{801} + R_{550}) \tag{4}$$

Also included in this group is a modification of the Chlorophyll Absorption in Reflectance Index (CARI) developed by Kim (1994) for minimizing the effects of nonphotosynthetic materials on spectral estimates of absorbed photosynthetically active radiation. The modified CARI (MCARI) is the depth of the chlorophyll absorption at 670 nm relative to the reflectance at 550 nm and 700 nm and is defined as shown in Eq. (5):

$$\mathrm{MCARI} = [(R_{700} - R_{670}) - 0.2(R_{700} - R_{550})\cdot(R_{700}/R_{670}) \tag{5}$$

The intrinsic indices are often very sensitive to background reflectance properties and are often difficult to interpret at low leaf area indices (Rondeaux et al., 1996).

Soil-line vegetation indices use coefficients based on the generally linear relationship between near-infrared and visible reflectance for bare soil to reduce the influence of the soil on canopy reflectance. The Soil-Adjusted Vegetation Index (SAVI) is less sensitive to soil reflectance at low LAI than NDVI. Huete (1988) defined SAVI as [see Eq. (6)]:

$$\mathrm{SAVI} = (1 + L)\cdot(R_{801} - R_{670})/(R_{801} + R_{670} + L) \tag{6}$$

where the constant $L = 0.5$ has been adjusted to account for first-order soil background variation. Numerous modifications of the SAVI concept have been reported (Baret

Table 3. Percent of Variation in Reflectance Factors and Spectral Vegetation Indices Associated with Background Reflectance (B), Chlorophyll Concentration (Chl), Leaf Area Index (LAI), and Their Interactions

	Source of Variation					
Spectral Variable	*Background*	*Chl*	*LAI*	*B·Chl*	*B·LAI*	*Chl·LAI*
R_{550}	33.1	16.6	0.4	–	43.7	6.2
R_{670}	16.7	–	58.3	–	25.0	–
R_{700}	26.8	5.7	30.2	–	35.3	2.0
R_{801}	8.0	0.1	87.7	–	4.0	0.2
NIR/Red	–	0.2	99.4	–	0.3	0.1
NIR/Green	0.7	12.3	80.0	–	0.9	6.1
NDVI	0.5	–	98.8	–	0.7	–
Green NDVI	2.5	6.0	85.5	–	4.1	1.9
MCARI	0.1	27.0	59.7	–	0.1	13.2
SAVI	0.4	–	98.9	–	0.7	–
OSAVI	0.5	–	98.8	–	0.7	–
Degrees of freedom	9	2	10	18	90	20

Percentages less than 0.1 are omitted for clarity.

and Guyot, 1991). Rondeaux et al. (1996) showed that the value of the L parameter is critical in the minimization of soil reflectance effects and proposed the Optimized SAVI (OSAVI) [see Eq. (7)]:

$$OSAVI=(1+0.16)\cdot(R_{801}-R_{670})/(R_{801}+R_{670}+0.16) \quad (7)$$

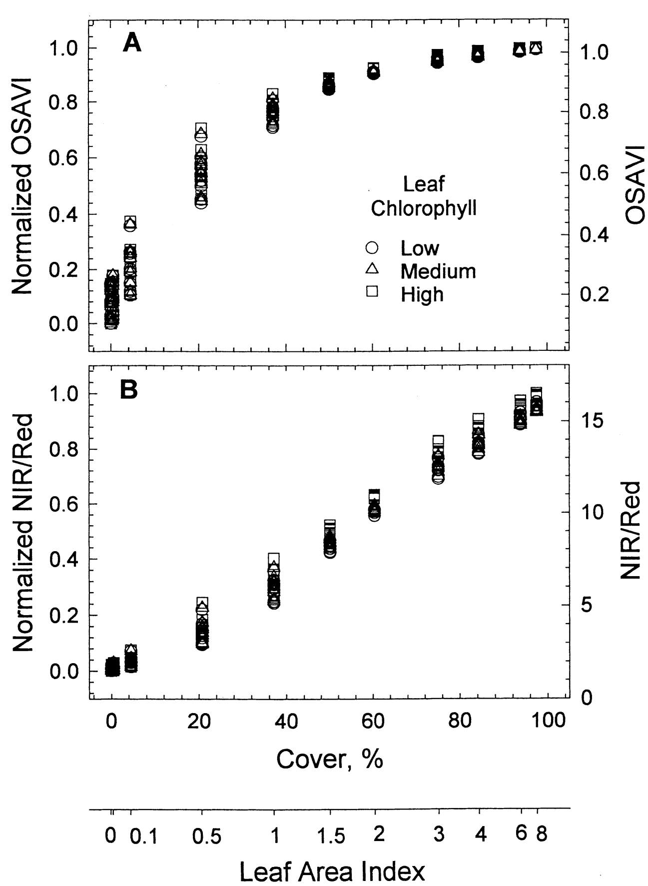
where L=0.16 was the optimized value.

All spectral vegetation indices were normalized between 0 for the minimum value and 1 for the maximum value. Analysis of variance (ANOVA) is useful for assessing what proportions of the variations in a dependent variable can be accounted for by one or more independent variables (Daughtry et al., 1980; Rondeaux et al., 1996). The impact of background reflectance, leaf chlorophyll, and LAI on selected reflectance factors and spectral vegetation indices are presented in Table 3. Background reflectance, LAI, and their interaction (B·LAI) accounted for significant proportions (77%–99%) of the variation in the single-band reflectance factors. For vegetation indices that used the near-infrared (R_{801}) and red (R_{670}) reflectance (i.e., NIR/Red, NDVI, SAVI, and OSAVI), LAI was the main variable accounting for >98% of the variation. Chlorophyll, LAI, and the interaction (Chl·LAI) accounted for >93% of the variation in the spectral indices that included a green band (R_{550}). Background reflectance accounted for less than 1% of the variation of each vegetation index, except Green NDVI, which was 2.5% (Table 3).

Rondeaux et al. (1996) reported similar results in their comparisons of the sensitivities of several vegetation indices to soil background effects, leaf angle distributions, and LAI. They concluded that OSAVI offered the best results for most agricultural crops, and they recommended its use for monitoring vegetation changes on seasonal or annual time scales. For the agricultural soils and crop residues used in the current study, OSAVI, SAVI, NDVI, and NIR/red were nearly equal in minimizing the contributions of background reflectance. The vegetation index of choice is based on its functional relationship with an agronomic characteristic of interest, generally LAI or foliage cover. Fig. 10 shows OSAVI and NIR/Red as a function of foliage cover and LAI. Foliage cover was defined as the percent of the soil surface covered by leaves and was calculated as shown in Eq. (8):

$$cover=100[1-\exp(-0.463\ LAI)] \quad (8)$$

Figure 10. Simulated vegetation indices plotted as a function of foliage cover and LAI for 10 backgrounds and three leaf chlorophyll levels: (A) OSAVI and (B) NIR/Red.



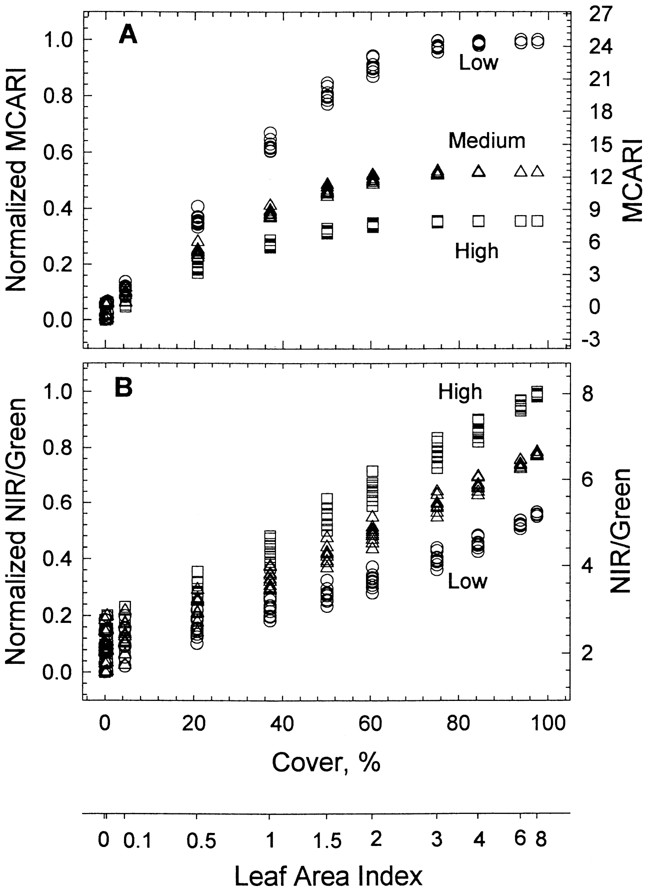


Figure 11. Simulated vegetation indices plotted as a function of foliage cover and LAI for 10 backgrounds and three leaf chlorophyll levels: (A) MCARI and (B) NIR/Green.

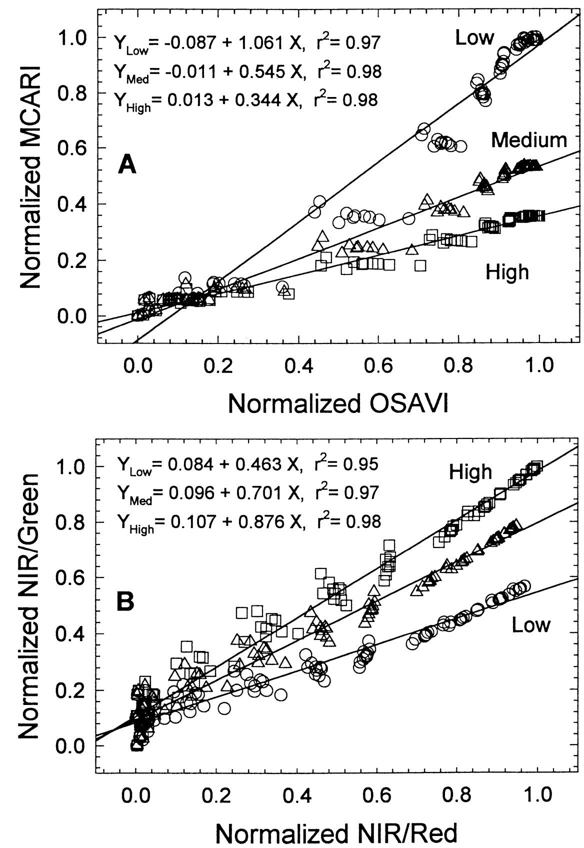


Figure 12. Plots of (A) MCARI vs. OSAVI and (B) NIR/Green vs. NIR/Red. Regression lines represent mean isolines of equal leaf chlorophyll concentrations.

where 0.463 is the extinction coefficient related to leaf angle distribution of corn (Daughtry et al., 1992). Vegetation indices are generally more linearly related to foliage cover than to LAI, especially at high values of LAI when the vegetation indices approach an asymptote (Rondeaux et al., 1996). In Fig. 10, as foliage cover (or LAI) increased, the range of OSAVI values decreased, particularly for foliage covers >40% (LAI>1.0). Thus, we can reliably estimate foliage cover (or LAI) with either of the vegetative indices in Fig. 10.

Unfortunately, the vegetation indices that are insensitive to background reflectance are also relatively insensitive to leaf chlorophyll concentration. The chlorophyll main effect and the chlorophyll·LAI interaction (Chl·LAI) accounted for 18.4% and 40.2% of the variation in NIR/green and MCARI, respectively (Table 3). For both of these vegetation indices, the three clusters of points at each foliage cover value are associated with differences in leaf chlorophyll concentration (Fig. 11). At each foliage cover, low values of NIR/Green indicate low leaf chlorophyll, while high values of MCARI indicate low chlorophyll.

In Fig. 12, MCARI was plotted as a function of OSAVI and NIR/Green as a function of NIR/Red. The three lines represent mean isolines of leaf chlorophyll concentrations. The coefficients of determination (r^2) for each mean line exceeded 0.95 with root mean square errors (RMSE) decreasing (Fig. 12A) or slightly increasing (Fig. 12B) as chlorophyll concentration increased. Background reflectance contributed to the scatter in each vegetation index, particularly at foliage covers <50%. When individual regression lines were fit for each background, average r^2 increased slightly (r^2>0.98) and RMSE decreased significantly (RMSE=0.033). Thus, customizing the relationships in Fig. 12 to a specific set of soils or crop residues could significantly improve remotely sensed estimates of leaf chlorophyll concentrations.

The three lines in Figs. 12A and 12B intersect near the bare soil values and radiate outward as foliage cover and chlorophyll density (i.e., product of LAI and chlorophyll concentration) increased. For NIR/Green vs. NIR/Red (Fig. 12B), the slopes of the three lines increased as leaf chlorophyll concentration increased. However, the slopes decreased as leaf chlorophyll increased for MCARI vs. OSAVI (Fig. 12A). In both Figs. 12A and 12B, one spectral vegetation index (OSAVI or NIR/Red) minimized the influence of background reflectance and leaf reflectance, while the second spectral vegetation index of each pair provided information on chlorophyll concentration. Other combinations of the spectral vegetation indices were plotted and evaluated, but none were as linear as those in Fig. 12.

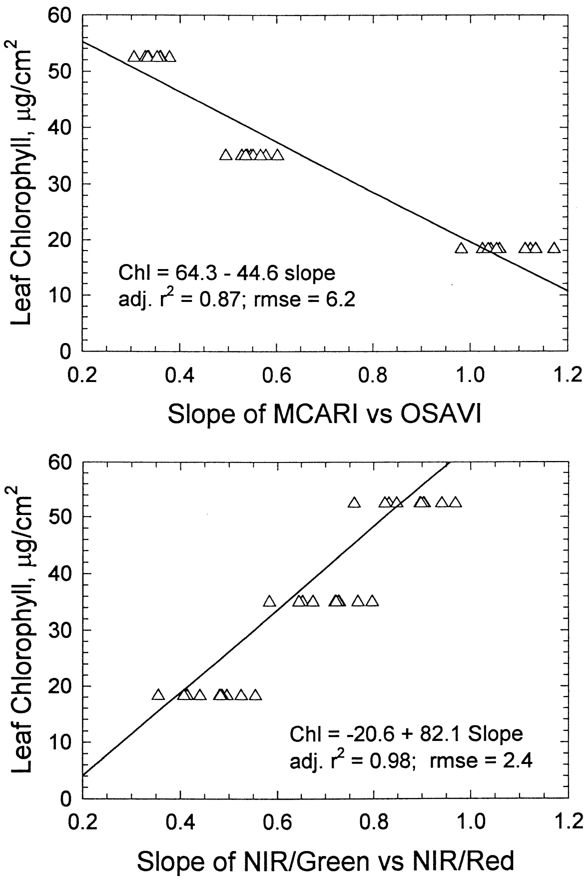


Figure 13. Leaf chlorophyll concentration as a function of the mean isoline slopes of (Top) MCARI vs. OSAVI and (Bottom) NIR/Green vs. NIR/Red from Figs. 12A and 12B, respectively. The 10 points at each leaf chlorophyll concentration represent the slopes of the vegetation indices for each background (five backgrounds·two moistures) used in the SAIL model.

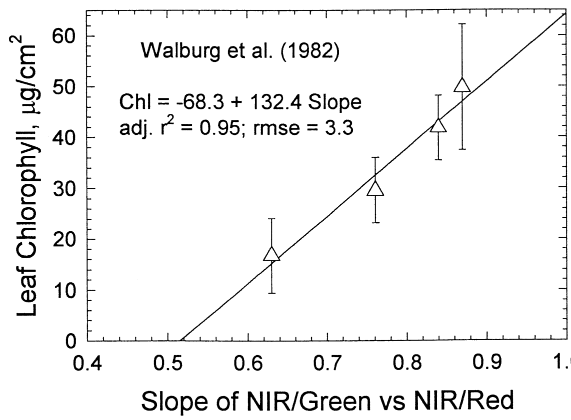


Figure 14. Measured leaf chlorophyll concentration as a function of isoline slopes for TM4/TM2 (i.e., NIR/Green) vs. TM4/TM3 [i.e., (NIR/Red) for measured canopy reflectance using data from Walburg et al. (1982)]. The error bars represent ±1 standard deviation of the measured chlorophyll values for each N rate.

Thus, leaf chlorophyll concentration is related to the slope of these pairs of spectral vegetation indices. In Fig. 13, leaf chlorophyll concentrations used in the SAIL model appear to be a linear function of slope of each regression isoline from Fig. 12. Although there are only three leaf chlorophyll concentrations, these simulated data span a wide range of foliage cover (or LAI) values and background reflectance values. Intermediate values of leaf chlorophyll could be estimated by calculating the slope of MCARI vs. OSAVI or NIR/Green vs. NIR/Red.

Measured Canopy Reflectance

As a limited test of the preceding analysis of simulated data, we used corn canopy reflectance data in Landsat TM bands from Walburg et al. (1982) to calculate the spectral vegetation indices in Eqs. (1) through (7). NIR/Green (i.e., TM4/TM2) was plotted as a function of NIR/Red (i.e., TM4/TM3) for the four N treatments on seven dates prior to physiologic maturity of the corn. As with the simulated data, mean leaf chlorophyll concentrations were a linear function of the slopes of the vegetation indices (Fig. 14).

SUMMARY

Leaf chlorophyll concentration is an indicator of plant N status. Changes in leaf chlorophyll concentrations produce rather broad-band differences in leaf reflectance and transmittance spectra. However, the transition from leaf spectra to canopy reflectance spectra is complicated. Variations in background reflectance and LAI confounded detection of relatively subtle differences in canopy reflectance due to changes in leaf chlorophyll concentration. Some spectral vegetation indices (e.g., OSAVI and NIR/Red) minimized background reflectance contributions, while other indices (e.g., MCARI and NIR/Green) responded more to leaf chlorophyll concentrations. Combining these two groups of spectral vegetation indices in the same figure produced isolines of leaf chlorophyll concentrations. The ratio of pairs of these spectral vegetation indices was linearly related to leaf chlorophyll concentration over a wide range of foliage cover and background reflectance. The current study used simulated data to demonstrate that pairs of spectral vegetation indices can estimate leaf chlorophyll concentrations with minimal confounding effects due to LAI and background reflectance. The effects of atmospheric conditions on these vegetation indices must be evaluated. Although we recognize that these relationships need to be examined further with real, not simulated, data, this was beyond the scope of the current study.

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