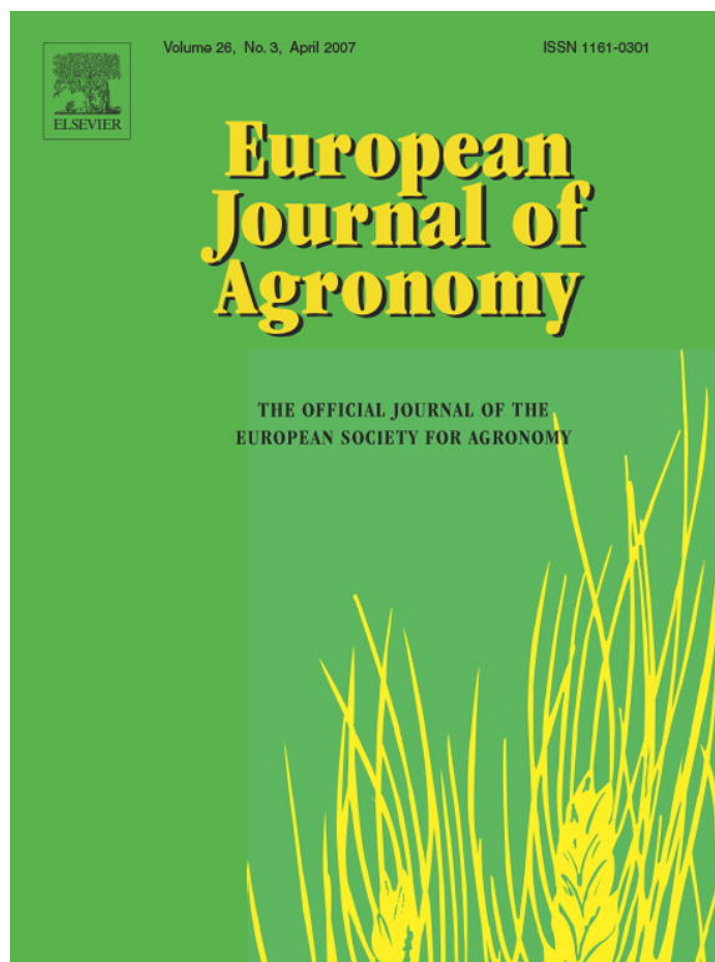


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## Canopy reflectance in cotton for growth assessment and lint yield prediction

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Received 5 June 2006; received in revised form 26 November 2006; accepted 19 December 2006

### Abstract

A field experiment was conducted in 2001–2002 to investigate relationships between canopy spectral reflectance and leaf area index (LAI), aboveground biomass (ABM), and lint yield of irrigated cotton across four N fertilizer rates of 0, 56, 112, and 168 kg N ha<sup>-1</sup>. These N rates were used to generate a wide range of difference in canopy structure and lint yield. Measurements of canopy reflectance were made throughout the growing season using a hand-held spectroradiometer. Samples for LAI and ABM were obtained four (2001) or five (2002) times during squaring and fruiting. Mean reflectance values in red ( $R_{red}$ ) and near infrared ( $R_{NIR}$ ) regions were obtained from canopy reflectance data based on the Landsat Thematic Mapper bands. The reflectance ratio vegetation index (RVI), normalized difference vegetation index (NDVI), enhanced vegetation index (EVI), wide dynamic range vegetation index (WDRVI) as well as several hyperspectral reflectance indices were calculated. Most reflectance indices had exponential relationships with both LAI and ABM and reached saturation at high LAI and ABM, but were linearly correlated with log(LAI) and log(ABM). Relative lint yield was linearly correlated to the reflectance indices measured any time after the first square stage and the strongest correlation was obtained at the early flower stage with  $r^2$  of 0.56–0.89 ( $P < 0.01$ ). Therefore, the canopy reflectance indices measured at early flower stage of cotton growth could serve as input to a crop growth model for predicting potential yield loss. These results indicate that the early flower stage is an appropriate time to collect canopy reflectance data for cotton yield estimation.

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**Keywords:** Cotton (*Gossypium hirsutum* L.); Leaf area index; Aboveground biomass; N fertilizer rate; Lint yield; Canopy reflectance; Remote sensing

### 1. Introduction

Cotton (*Gossypium hirsutum* L.) is responsive to changes in growth environment due to its indeterminate growth habit (Reddy et al., 1997). Nitrogen (N) management is one of the important practices in high-yielding cotton production systems (Gerik et al., 1998). Both N deficiency and excess N negatively affect plant growth, boll retention, lint yield, and fiber quality (Gerik et al., 1998; Reddy et al., 2004). Insufficient N supply often results in smaller leaf area (Fernandez et al., 1996; Zhao and Oosterhuis, 2000) and lower leaf photosynthesis and biomass production (Zhao and Oosterhuis, 2000), resulting in reduced lint yield and poor fiber quality (Heagle et al., 1999; Reddy et al., 2004). However, lint yield of irrigated cotton does

not always continue to increase as amount of N fertilizer is increased (Wood et al., 1992; Boquet et al., 1994). When N rate reaches a certain amount, a further increase in N fertilizer may limit lint yield if fruit abscission is increased due to poor light environment in the canopy. Excess use of N fertilizer increases not only production cost but also the potential for environmental problems, such as groundwater contamination (Jaynes et al., 2001).

Remotely sensed data have been widely used to develop vegetation indices as indicators of crop growth, nutrient status and yield development. Studies suggest that crop spectral reflectance can be used to detect abiotic and biotic environmental stresses (Filella et al., 1995; Osborne et al., 2002), assess plant nutrient status (Zhao et al., 2005a), estimate plant growth and physiology (Peñuelas and Filella, 1998; Serrano et al., 2000; Broge and Leblanc, 2001; Zhao et al., 2003), monitor plant conditions at various scales (Blackmer et al., 1994; Plant et al., 2001), and predict crop yields (Blackmer et al., 1994; Plant et al., 2001; Ma et al., 2001). Two broad band reflectance

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indices of near infrared (NIR) to red ratio vegetation index (RVI) and normalized difference vegetation index (NDVI) have been widely used to assess ground coverage of plant vegetation, leaf area index (LAI), biomass production and crop economic yields. Studies have found that RVI and/or NDVI asymptotically saturate in high biomass or great LAI (Huete et al., 2002; Gitelson, 2004). Similarly, Reddy et al. (2003) investigated relationships of cotton LAI and aboveground biomass (ABM) with several canopy hyperspectral reflectance indices and reported that saturation of the vegetation indices still existed in higher LAI or ABM. It is reported that broad band reflectance indices of EVI or WDRVI can minimize the saturation problem (Gitelson, 2004). Many hyperspectral reflectance indices, such as chlorophyll absorption reflectance index (CARI), modified CARI (MCARI), transformed CARI (TCARI), optimized soil-adjusted vegetation index (OSAVI), TCARI/OSAVI (Daughtry et al., 2000; Haboudane et al., 2002), have been developed for plant canopy LAI and chlorophyll content assessment. More recently, Haboudane et al. (2004) designed several other hyperspectral reflectance indices and reported that the most robust indices were modified triangular vegetation index (MTVI2) and modified chlorophyll absorption reflectance index (MCARI2). These two indices were less sensitive to chlorophyll content variations and linearly related to green LAI (Haboudane et al., 2004). It has been reported that the first derivative of red edge reflectance ( $dR/d\lambda$ ) at 740 nm is highly related to leaf N or chlorophyll content (Zhao et al., 2005b). Although numerous reflectance indices have been developed from leaves and canopy levels of various plants, limited information is available in determining if these hyperspectral reflectance indices can be used to assess cotton growth and yield parameters.

Many studies have documented the relationships between crop yields and broad band indices, RVI and NDVI (Aparicio et al., 2000; Serrano et al., 2000; Shanahan et al., 2001; Ma et al., 2001; Plant et al., 2001; Dobermann and Ping, 2004; Zarco-Tejada et al., 2005). Benedetti and Rossini (1993) used NDVI calculated from satellite images to predict grain yield in wheat (*Triticum aestivum* L.). Ma et al. (2001) reported that yield of different soybean (*Glycine max* L.) genotypes was closely correlated with canopy NDVI measured around the pod-filling stage. Li et al. (2001) found a close correlation between cotton yield and NDVI measurements made during fruiting using a hand-held spectroradiometer at 2 m above the canopy. On the other hand, some studies have found no significant relationships between NDVI and crop yield (Pettigrove et al., 1999; Plant et al., 1999; Bronson et al., 2005). Most recently, Zarco-Tejada et al. (2005) investigated temporal and spatial relationships between cotton yield variability and many hyperspectral reflectance indices and concluded that the relationships depended on the time of image acquisition. Yield in cotton is correlated with the amount of photosynthetic tissue, which can be estimated from reflectance in the NIR and red regions of the spectrum as it is associated with changes in vegetation ground coverage or LAI (Plant et al., 2001). If cotton plant growth and lint yield could be predicted using the canopy reflectance indices at early- and mid-growing season, it would help growers make field management deci-

sions such as irrigation, foliar N and plant growth regulator applications for maximum yield.

Nitrogen supply directly or indirectly affects LAI, canopy coverage, chlorophyll content, and other biophysical parameters, which may result in changes in canopy reflectance indices of RVI, NDVI, EVI, WDRVI, TCARI, MCARI, OSAVI, TCARI/OSAVI, MCARI/OSAVI, MTVI2, MCARI2, and the first derivative of red edge reflectance in 740 nm ( $dR/d\lambda$ ). The objective of this study was to determine relationships between these reflectance indices of irrigated cotton and LAI, ABM, and lint yield across different N fertilizer rates. Our results should provide new approaches for predicting cotton biophysical, growth, and lint yield parameters from remotely sensed data based on the radiative transfer accounting for canopy structure and leaf optical properties.

## 2. Materials and methods

### 2.1. Plant culture

The experiment was conducted in the 2001 and 2002 growing seasons on a Leeper silt clay loam (Fine, smectitic, nonacid, thermic Vertic Epiaquept) at the Mississippi Agricultural and Forestry Experiment Station (33°28'N, 88°47'W), Mississippi State University, MS, USA. Nitrate-N content in top 50-cm soil was determined prior to planting in both the years and was 32.5 kg ha<sup>-1</sup> in 2001 and 22.4 kg ha<sup>-1</sup> in 2002. Seeds of cotton cv. NUCOTN 33B, a mid season maturity cultivar, were seeded on 14 May 2001 and 24 May 2002. Rows were spaced 1 m apart and oriented in an east-west direction. Seedlings were thinned to a density of about nine plants per meter row when plants reached the second true-leaf stage.

### 2.2. Treatments

To generate a wide range of crop canopy characteristics and final lint yield, plots were fertilized with 0, 56, 112, and 168 kg N ha<sup>-1</sup> applied as a liquid fertilizer of N solution and suspensions (32% N) injected beside each row. These treatments were: (1) no N applied during the growing season (0N); (2) 56 kg N ha<sup>-1</sup> applied at the second true leaf stage (56N); (3) 112 kg N ha<sup>-1</sup>, equally split (56 and 56 kg ha<sup>-1</sup>) and applied at the second true leaf stage and at the first square (FS) stage (112N); (4) 168 kg N ha<sup>-1</sup> split into two applications of 56 and 112 kg N ha<sup>-1</sup> and applied at the second true leaf stage and at the FS stage (168N, control), respectively. Except for the N rates, all plots received the same field management practices, such as irrigation, P and K fertilizer applications, and insect and weed control. These field management practices were based on current Mississippi cotton production recommendations. The experiment was arranged in a randomized complete block design with five (2001) or six (2002) replications. Plot size was 8 m wide by 15 m long with eight rows per plot in both the years.

### 2.3. Measurements

Plant growth stage of FS was defined as the date when 50% of plants had a first-visible floral bud (square) with a 3-mm

dimension and first flower (FF) stage was defined as the date when 50% of plants had the first white flower. Dates of the FS and FF in all the treatments in 2001 were 23 June [40 days after seeding (DAS)] and 17 July (64 DAS), respectively and in 2002 were 28 June (35 DAS) and 19 July (56 DAS), respectively.

Starting from 42 (2001) or 22 DAS (2002) through boll opening, canopy reflectance measurements were made on sunny days between 1100 and 1300 h above all plots using a portable ASD FieldSpec FR spectroradiometer (Analytical Spectral Devices Inc., Boulder, CO, USA<sup>1</sup>). Reflectance was measured at wavelengths ranging from 350 to 2500 nm with a 1-nm sampling interval. The distance between the optical head of the spectroradiometer and the plant terminal was 2 m throughout the season. The radiometer had a 25° field of view. After optimization of the ASD instrument, a Spectralon (Labsphere, Inc., Sutton, NH, USA) white panel was used to obtain reference signal prior to taking three canopy reflectance measurements from each plot. The canopy reflectances were computed as the ratio of canopy radiances to the radiances from the white reference panel.

In order to determine relationships between reflectance and plant growth, all of the plants in a randomly selected 1-m length of row from each plot were harvested at four to five time periods: 43, 52, 67, and 84 DAS in 2001 and 34, 45, 52, 69, and 83 DAS in 2002. Canopy reflectance was also measured as described above around these specific dates. Harvested plants were transported to the laboratory and separated into leaves, stems and fruits. Green leaf area was measured using a LI-3100 leaf area meter (LI-COR Inc., Lincoln, NE, USA). Plant tissues were dried in a forced-air oven at 70 °C for 72 h and weighed. Values for LAI and ABM were further calculated from these laboratory measurements.

When approximately 60% of bolls opened in the 112-N treatment plots (25 September 2001 and 28 September 2002), all plots were defoliated with a mix of commercial defoliation chemicals of DROP (active ingredient: *N*-phenyl-*N*,1,2,3-thiadiazol-5-ylurea), FOLEX (*s,s,s*-tributyl phosphorothioate) and PREP (ethephon phosphinic acid) based on Mississippi State University Cotton Defoliation Recommendation (<http://msucares.com/pubs/infosheets/is0529.pdf>). Seed cotton in the middle four rows of each plot was harvested mechanically about 2 weeks after defoliation in 2001. In 2002, the harvest date was delayed to 3 December due to abnormally high precipitation between September and November. Therefore, the harvested yield might underestimate the actual yield due to boll rot caused by extreme wet weather during the post defoliation period in 2002. A sub-sample of 500-g seedcotton from each plot was collected and ginned to calculate lint percentage (*i.e.* lint weight/seedcotton weight × 100). Lint yields (kg ha<sup>-1</sup>) were obtained by multiplying the seedcotton yield by lint percentage. In order to minimize the effect of abnormal wet weather prior to harvest in 2002 and to make comparison between years, relative lint yield, defined as percentage of the maximum, was used for regression analysis of yield and the reflectance indices.

Relative lint yield was calculated by dividing the lint yield of each plot by the maximum lint yield (1671 kg ha<sup>-1</sup> in 2001 and 1498 kg ha<sup>-1</sup> in 2002) within a year.

#### 2.4. Data analysis

The three spectral reflectance measurements in each plot at each sampling date were averaged and the mean values were used in statistical analysis. Reflectance values in four-wavelength ranges (*i.e.* 350–399, 1350–1449, 1750–1969, and 2350–2500 nm) were omitted from data analysis due to noise and location of these bands within regions of atmospheric water absorption. From the remaining reflectance data, hyperspectral reflectance indices, TCARI, MCARI, OSAVI, MTVI2, MCARI2, and  $dR/d\lambda$  were calculated based on the equations provided by Daughtry et al. (2000), Haboudane et al. (2002), Haboudane et al. (2004) and Zhao et al. (2005a,b):

TCARI

$$= 3 \times \left[ (R_{800} - R_{670}) - 0.2 \times (R_{800} - R_{550}) \times \left( \frac{R_{700}}{R_{670}} \right) \right],$$

$$\text{MCARI} = [(R_{700} - R_{670}) - 0.2 \times (R_{700} - R_{550})] \times \left( \frac{R_{700}}{R_{670}} \right),$$

$$\text{OSAVI} = (1 + 0.16) \times \left( \frac{(R_{800} - R_{670})}{(R_{800} + R_{670} + 0.16)} \right),$$

$$\text{MTVI2} = \frac{1.5 \times [1.2 \times (R_{800} - R_{550}) - 2.5 \times (R_{670} - R_{550})]}{\sqrt{(2 \times R_{800} + 1)^2 - (6 \times R_{800} - 5 \times \sqrt{R_{670}}) - 0.5}},$$

$$\text{MCARI2} = \frac{1.5 \times [2.5 \times (R_{800} - R_{670}) - 1.3 \times (R_{800} - R_{550})]}{\sqrt{(2 \times R_{800} + 1)^2 - (6 \times R_{800} - 5 \times \sqrt{R_{680}}) - 0.5}},$$

$$\frac{dR}{d\lambda} = \frac{R_{740} - R_{730}}{10}$$

where  $R_{550}$ ,  $R_{670}$ ,  $R_{680}$ ,  $R_{700}$ ,  $R_{730}$ ,  $R_{740}$  and  $R_{800}$  represent reflectance at 550, 670, 680, 700, 730, 740, and 800 nm, respectively. In addition, six broad wavebands were selected based on Landsat Thematic Mapper (TM). These individual wavebands were in the blue (450–520 nm), green (520–600 nm), red (630–690 nm), NIR (760–900 nm), short-wave infrared 1 (SWIR1, 1550–1750 nm), and short-wave infrared 2 (SWIR2, 2080–2350 nm) regions of the spectrum. Four broad-band reflectance indices, RVI, NDVI, EVI, and WDRVI, were calculated using the following formulae according to Rouse et al. (1974), Huete et al. (2002), and Gitelson (2004):

$$\text{RVI} = \frac{R_{\text{NIR}}}{R_{\text{red}}}, \quad \text{NDVI} = \frac{R_{\text{NIR}} - R_{\text{red}}}{R_{\text{NIR}} + R_{\text{red}}},$$

$$\text{EVI} = 2.5 \times \frac{R_{\text{NIR}} - R_{\text{red}}}{R_{\text{NIR}} + 6 \times R_{\text{red}} - 7.5 \times R_{\text{blue}} + 1},$$

$$\text{WDRVI} = \frac{0.2 \times R_{\text{NIR}} - R_{\text{red}}}{0.2 \times R_{\text{NIR}} + R_{\text{red}}}$$

where  $R_{\text{blue}}$ ,  $R_{\text{red}}$ , and  $R_{\text{NIR}}$  represent canopy reflectance in blue (450–520 nm), red (630–690 nm) and NIR (760–900 nm) regions, respectively. The mean LAI and ABM, as well as values of all the reflectance indices, for each treatment and sampling

<sup>1</sup> Use of trade or product names is for informational purpose only and does not imply endorsement by the United States Department of Agriculture to the exclusion of any other product that may be suitable.

date were obtained by averaging the data across replications to determine seasonal patterns of plant growth and reflectance indices as affected by N fertilizer rates.

One-way ANOVA and statistical tests of least significant differences (LSD) were employed to determine effects of N fertilizer rate on reflectance indices, plant growth, and relative yield (SAS Institute Inc., 1997). Values for LAI, ABM, and the reflectance indices were pooled over plots, sampling dates, and years to determine relationships between reflectance indices and LAI or ABM. Log transformations were used for LAI and ABM in an effort to linearize. Linear regression of the reflectance indices with  $\log(\text{LAI})$  and  $\log(\text{ABM})$  was then carried out. Furthermore, the reflectance data on each measuring date were pooled over the experimental plots and Pearson correlation coefficients ( $r$ ) were calculated between relative lint yield and canopy reflectance at each wide waveband and the reflectance indices at different growth stages. Best growth stage for collecting canopy reflectance for yield estimation was determined based on the  $r^2$  values of the linear regression equations.

### 3. Results

#### 3.1. Temperature and precipitation

Annual mean temperatures in 2001 (17.3 °C) and in 2002 (17.2 °C) at the experimental location were similar to the long-term average (16.9 °C) over 28 years (1973–2000). Especially during plant growth from April to August in both 2001 and 2002, the monthly mean temperatures were very similar between the years and comparable to the long-term average (Fig. 1A). Total annual precipitation was 1536 mm in 2001 and 1530 mm in 2002, which is about 6% higher than the 28-year average of 1444 mm (Fig. 1B). Although total amounts of precipitation in both the years were very close to the long-term average, the distribution of rainfall among months of the growing season varied greatly between the 2 years and from the long-term average. In-furrow irrigation during cotton growth (June–August) was scheduled to eliminate water deficit effects, but excessive and frequent precipitation during September and November in 2002 caused some bolls to rot in all the plots.

#### 3.2. LAI, ABM and lint yield

Both LAI and ABM increased rapidly as plants progressed through squaring and fruiting (Fig. 2). The 56 and 112N treatments did not differ from the 168N treatment (control) in either LAI or ABM at any sampling date. After FF stage, LAI and ABM of the 0N treatment were significantly ( $P < 0.05$ ) less than controls provided with 168 kg N ha<sup>-1</sup>. On the final sampling date at 84 DAS in 2001 (83 DAS in 2002), the 0N treatment had 24% (52%) lower LAI and 20% (45%) less ABM as compared with the control.

Both N rate and year significantly ( $P < 0.001$ ) affected absolute lint yield and the interactive effect of N  $\times$  year on yield also was significant ( $P < 0.001$ ). When a comparison was made among N treatments within a year, lint yield did not differ in 2001 (Table 1). In 2002, however, lint yields of the 0 and 56N

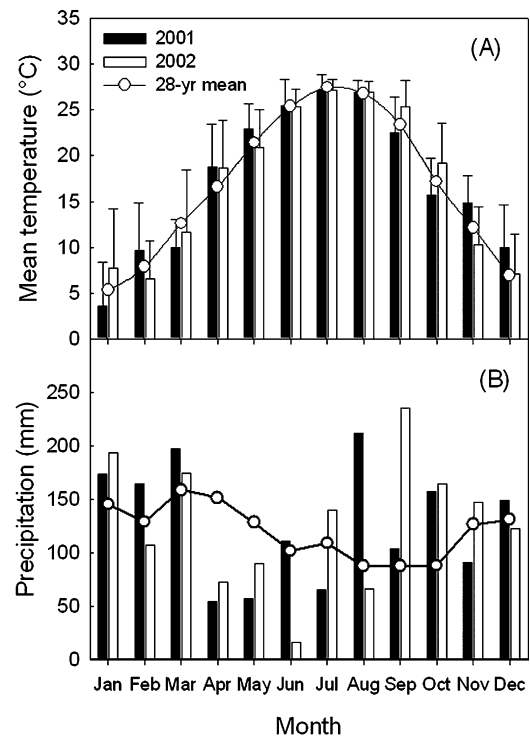


Fig. 1. Long-term (1973–2002) and (A) average monthly air temperatures + 1S.D. and (B) total monthly precipitation at the experimental location in 2001 and 2002.

treatments were 39 and 17% ( $P < 0.05$ ) less than that of the control of 168 kg N ha<sup>-1</sup> and lint yield of the 112N treatment did not differ from the control. Although lint yield did not differ statistically among the four N-rate treatments in 2001, the yield

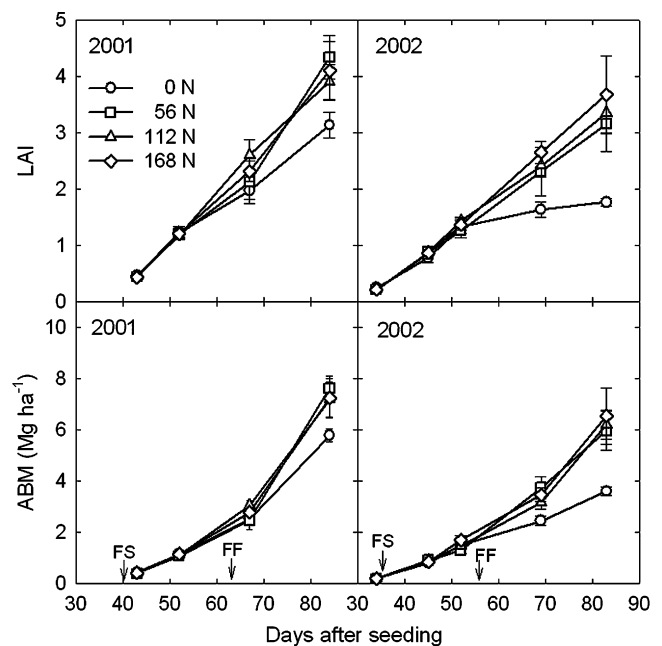


Fig. 2. Cotton leaf area index (LAI) and aboveground biomass (ABM) during squaring and fruiting as affected by N fertilizer treatments in 2001 and 2002. Each data point is the mean  $\pm$  S.E. of five (2001) or six (2002) replications. First square (FS) and first flower (FF) stages are also shown in the figure.

Table 1

Statistics for cotton absolute lint yield and relative lint yield, calculated by dividing the individual plot lint yield by the maximum yield  $\times 100$ , under four N fertilizer rate treatments in 2001 ( $n=20$ ) and 2002 ( $n=24$ )

Treatment	2001		2002	
	Absolute yield (kg ha <sup>-1</sup> )	Relative yield (%)	Absolute yield (kg ha <sup>-1</sup> )	Relative yield (%)
0N	1369 a	81.8 a	864 c	57.7 c
56N	1411 a	84.3 a	1190 b	79.4 b
112N	1464 a	87.5 a	1367 a	91.3 a
168N	1450 a	86.7 a	1426 a	95.2 a
Maximum	1671	100	1498	100
Minimum	1214	72.8	786	47.8
CV (%)	10	9.4	21	20.9

Treatment means within a column followed by the same letter are not significant ( $P>0.05$ ).

variability among plots existed (CV = 10%). In 2002, lint yield had much greater variation (CV = 21%) among 24 plots across treatments and replicates and ranged from 786 to 1498 kg ha<sup>-1</sup> (Table 1). Greater variability in lint yield across plots allowed us to determine the functional relationships between lint yield and canopy reflectance indices.

### 3.3. Reflectance indices

In early growing season, RVI, NDVI, EVI, WDRVI, OSAVI, MTVI2, MCARI2, and  $dR/d\lambda$  increased rapidly with the progression of plant growth, and reached maximum values about 1–2 weeks after the FF stage, while changes in TCARI, MCARI, TCARI/OSAVI, and MCARI/OSAVI during plant growth did not follow the same pattern (Fig. 3). Analyses of variances indicated that both N fertilizer rate and year significantly affected the reflectance indices around the early flower stage and the interactive effects of N  $\times$  year on the reflectance indices were also detected ( $P<0.001$ ).

In 2001, the 0N treatment had significantly smaller values for RVI, MTVI2, MCARI2 and  $dR/d\lambda$  and greater values for TCARI, MCARI and MCARI/OSAVI than the control (168N treatment) at 71, 78 and 100 DAS; otherwise, treatment differences were not significant (Fig. 3). In 2002, treatment differences in the canopy reflectance indices were detected from the FF stage onward. Low N treatments had greater TCARI, MCARI, TCARI/OSAVI, and MCARI/OSAVI and less other reflectance indices than the high N treatment. During flowering and fruiting in the 2002 growing season, the 112N treatment did not differ from the control in RVI, but the 0 and 56N treatments had 42 and 15% lower RVI, respectively, than the control when averaged across the five measuring dates between 55 and 102 DAS. The treatment differences in RVI and  $dR/d\lambda$  were much greater than those in all other indices (Fig. 3).

### 3.4. Relationships between LAI and ABM and reflectance indices

Relationships of cotton LAI with RVI, NDVI, EVI, WDRVI, OSAVI, MTVI2, MCARI2, and  $dR/d\lambda$  followed hyperbolic trends (Fig. 4). These reflectance indices increased linearly

with the increase in LAI when plants were relatively small. However, the indices did not respond to increases in LAI when LAI was  $>2$ . Relationships between ABM and these reflectance indices are similar to that of LAI and the indices did not respond to increase in ABM when ABM was  $>3$  Mg ha<sup>-1</sup> (data not shown). Furthermore, NDVI and OSAVI showed more saturation than other reflectance indices. Plots of  $\log(\text{LAI})$  versus these reflectance indices showed strong linear associations ( $r^2=0.64\text{--}0.75$ ,  $P<0.001$ ) (Fig. 4). Among the reflectance indices, WDRVI and OSAVI had the greatest  $r^2$  with  $\log(\text{LAI})$  and  $\log(\text{ABM})$ . The correlation of LAI (or ABM) with any of TCARI, MCARI, TCARI/OSAVI or MCARI/OSAVI was poor (Fig. 4).

### 3.5. Relationships between lint yield and reflectance indices

Analysis of the relationships between relative lint yield and the tested reflectance indices at different growth stages indicated that relative yield did not correlate with any other reflectance indices measured before 45 DAS except for TCARI/OSAVI, while  $r$  values between relative lint yield and the reflectance indices measured between 50 and 100 DAS were significant (Fig. 5). Moreover, the relative yield was most highly correlated with canopy reflectance indices (except for MTVI2 and MCARI2 in 2001, TCARI and MCARI/OSAVI in 2002) soon after FF stage was reached, 71 DAS in 2001 and 73 DAS in 2002. Relative lint yield negatively correlated with TCARI, MCARI, TCARI/OSAVI, and MCARI/OSAVI and positively correlated with other reflectance indices tested. TCARI/OSAVI seemed to be better correlated with relative yield compared to other indices in early growing season (around FS stage). Compared to broad band reflectance indices, hyperspectral indices tested in this study did not improve the relationships between cotton yield and remotely sensed data (Fig. 5).

Scatter plots of relative yield ( $Y$ ) versus RVI, NDVI, EVI, or WDRVI ( $X$ ) at different growth stages after FS indicated that the relationships could be expressed as a linear function (*i.e.*  $Y=a+bX$ ). Table 2 shows the regression equation parameters and  $r^2$  values of relative yield and RVI or NDVI generated for 2001 and 2002 at each sampling date. Linear equations developed after 45 DAS in the both years were highly significant ( $P<0.01$ ). The linear regression equations of relative

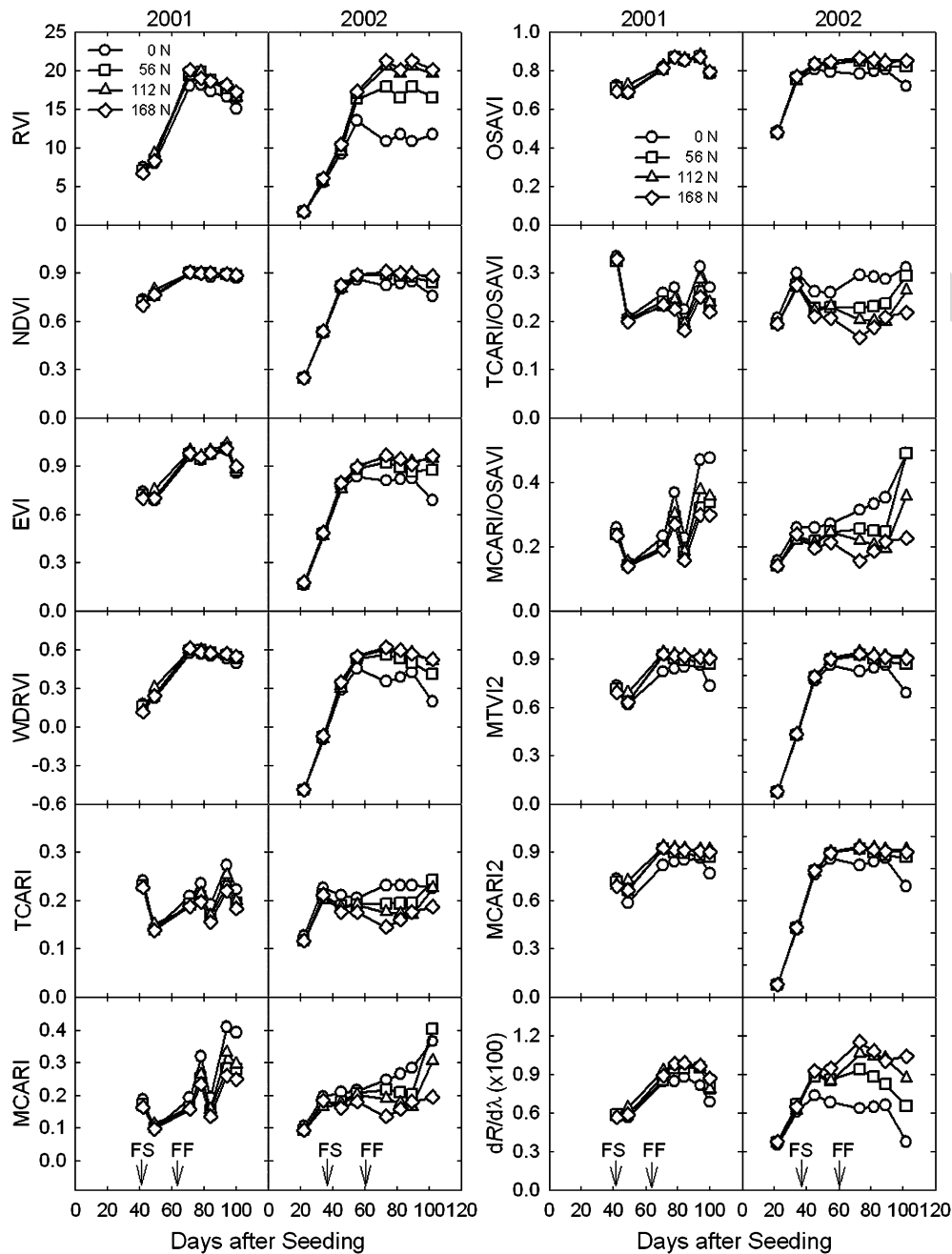


Fig. 3. Seasonal patterns of RVI, NDVI, EVI, WDRVI, TCARI, MCARI, OSAVI, TCARI/OSAVI, MCARI/OSAVI, MTVI2, MCARI2, and  $dR/d\lambda$  as affected by N fertilizer treatments. First square (FS) and first flower (FF) stages are shown in the figure.

lint yield with EVI, and WDRVI in early flower stage (71 DAS in 2001 and 73 DAS in 2002) were  $Y = -219.4 + 310.0X$  ( $r^2 = 0.57$ ) and  $Y = -64.0 + 158.3X$  ( $r^2 = 0.60$ ), respectively in 2001 and  $Y = -76.6 + 271.3X$  ( $r^2 = 0.62$ ) and  $Y = 11.7 + 129.3X$  ( $r^2 = 0.87$ ), respectively in 2002.

In order to test the feasibility of the linear models in early flower stage for relative lint yield estimation, the equations, developed from the reflectance data collected around the FF stage in each year, were used to predict relative lint yield of the other year. Scatter plots were used to further compare predicted and measured yields (Fig. 6). Among the four reflectance index models, the RVI model provided the best prediction of

the 2002 relative yield ( $r^2 = 0.87$ ) with a root-mean-square error (RMSE) of 7.7%. When the 2001 NDVI and WDRVI models were validated, the three lowest yield plots were considerably underpredicted (Fig. 6A). Removal of these three outliers from the validation scatter plot gave a much improved fit with  $r^2$  of 0.75 and RMSE of 8.1% for the NDVI model and with  $r^2$  of 0.79 and RMSE of 7.8% for the WDRVI. When the reflectance models developed in 2002 were used to predict the 2001 yield, the output results of RVI and NDVI models were poor compared to the 2001 models (Fig. 6B). The models underestimated relative yield when it was higher than 85%. Otherwise, the yield was overestimated.

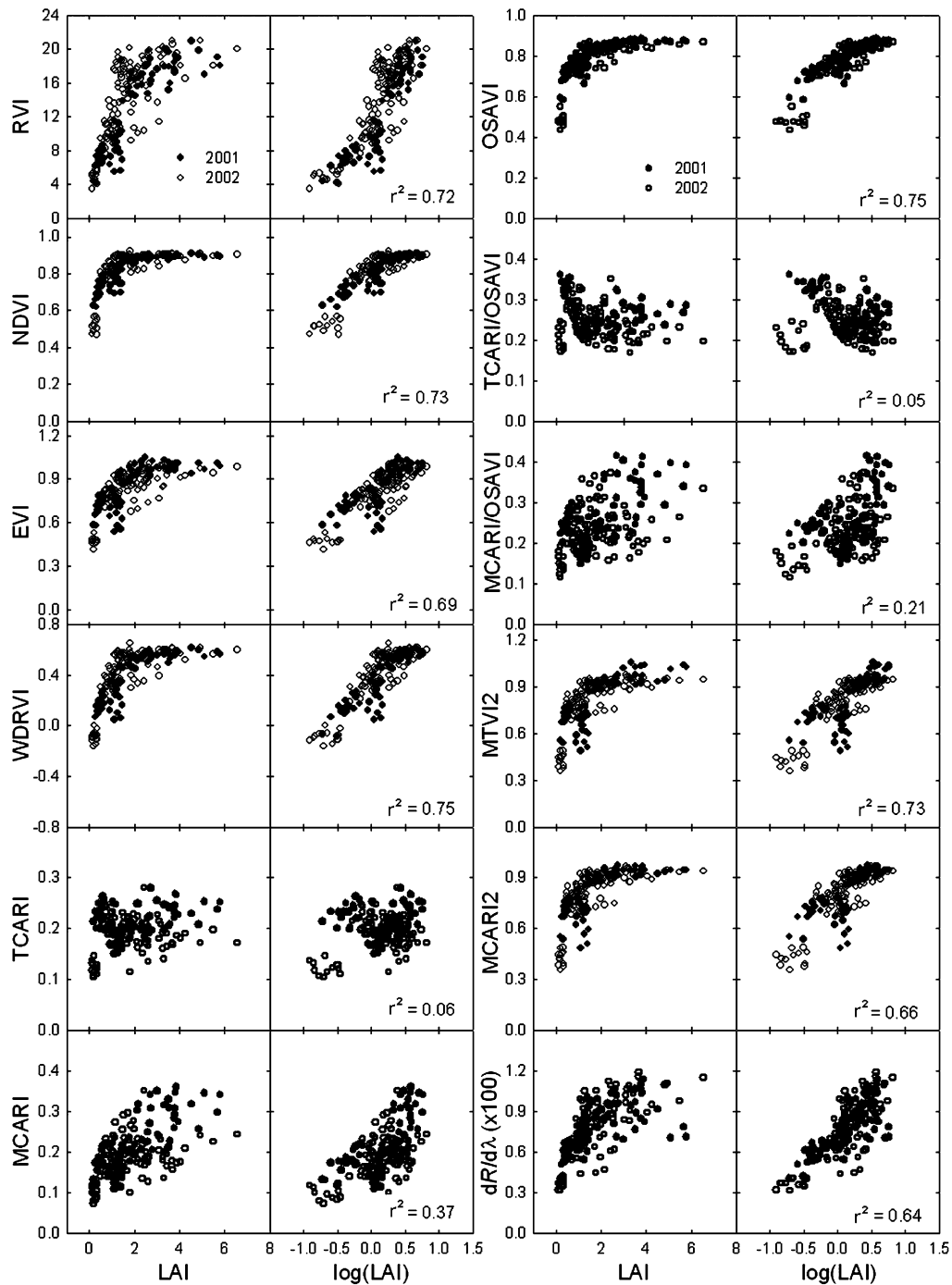


Fig. 4. Relationships of cotton leaf area index (LAI) and aboveground biomass (ABM) with RVI, NDVI, EVI, WDRVI, TCARI, MCARI, OSAVI, TCARI/OSAVI, MCARI/OSAVI, MTVI2, MCARI2, and  $dR/d\lambda$ . Data represent values for individual plots pooled across N treatments, replicate plots, sampling dates, and years ( $n=200$ ).

#### 4. Discussions

Although cotton growth responds well to N supply (Reddy et al., 1997; Gerik et al., 1998), lint yield of cotton does not always increase as amount of N application rate is increased (Wood et al., 1992; Boquet et al., 1994). A lack of differences among the N treatments in lint yield in 2001 (Table 1) was probably associated with a high initial level of soil N content. This is supported from nitrate-N content of  $32.5 \text{ kg ha}^{-1}$  in 2001 and

$22.4 \text{ kg ha}^{-1}$  in 2002 in the top 50-cm soil samples tested at the seeding dates in the present study. Because cotton yield response to N application rates depends on initial soil fertility and other environmental factors (Gerik et al., 1998; Fritsch et al., 2003), the relationship between cotton yield and N fertilizer rate may not be linear. When the N application rate reaches an optimum amount, a further increase in N fertilization does not always improve cotton yield but may even decrease yield (Wood et al., 1992; Boquet et al., 1994) due to increases in



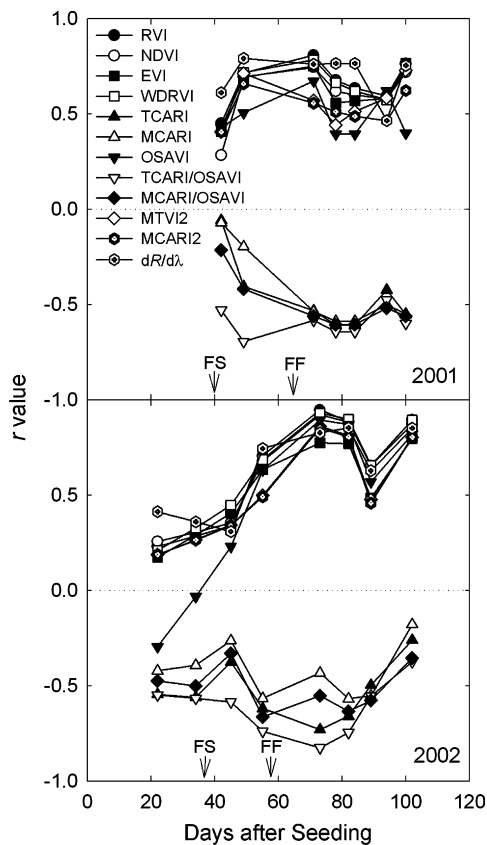


Fig. 5. Simple correlation coefficient ( $r$ ) between cotton relative lint yield and RVI, NDVI, EVI, WDRVI, TCARI, MCARI, OSAVI, TCARI/OSAVI, MCARI/OSAVI, MTVI2, MCARI2, and  $dR/d\lambda$  at different growth stages. The significant level of  $|r|$  values at  $P=0.05$  are 0.421 in 2001 ( $n=20$ ) and 0.397 in 2002 ( $n=24$ ). First square (FS) and first flower (FF) stages are also shown in the figure.

Table 2

Statistics for linear regression functions [ $Y=a+b(\text{RVI or NDVI})$ ] of cotton relative lint yield ( $Y$ ) in 2001 ( $n=20$ ) and 2002 ( $n=24$ ) with RVI or NDVI measured at different growth stages across four rates of N fertilizer

Year	DAS	RVI			NDVI		
		$a$	$b$	$r^2$	$a$	$b$	$r^2$
2001	42	65.55	2.77	0.171	55.70	41.07	0.081
	49	60.95	2.84	0.513***	11.45	95.52	0.483***
	71	-9.61	4.92	0.654***	-641.59	804.15	0.561***
	78	6.08	4.12	0.461**	-658.51	825.95	0.456**
	94	30.81	3.10	0.354**	-365.91	506.31	0.331**
2002	101	32.21	3.23	0.585***	-305.63	442.41	0.540***
2002	22	79.32	9.35	0.053	85.44	38.34	0.067
	45	44.37	3.72	0.115	-88.95	209.37	0.125
	53	-8.88	5.61	0.498***	-517.92	680.19	0.461***
	73	16.01	3.66	0.894***	-311.11	442.78	0.848***
	82	6.58	4.40	0.785***	-453.56	608.02	0.781***
	89	32.72	2.95	0.426***	-266.42	395.10	0.430***
2002	102	27.55	4.16	0.821***	-125.17	245.11	0.760***

DAS: days after seeding; RVI: ratios of reflectance at near infrared (NIR) to red (R); NDVI: normalized difference vegetation index. \*\* and \*\*\* indicate significance of  $r^2$  at  $P<0.01$  and 0.001 levels, respectively.

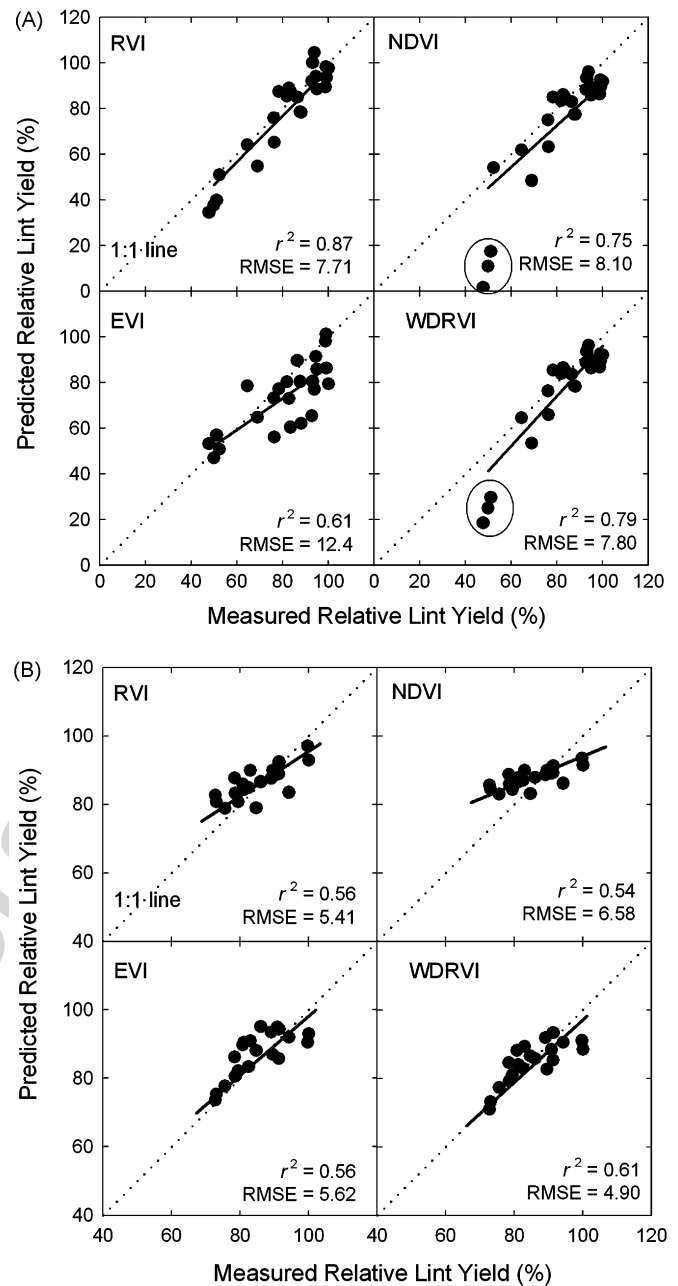


Fig. 6. Relationship between measured and predicted relative lint yield in (A) 2002 and (B) 2001 using linear regression models developed in 2001 and 2002, respectively (see figure) with RVI, NDVI, EVI, and WDRVI at early flower stage. The  $r^2$  and root-mean-square error (RMSE) values are also presented.

insect and disease pressure and boll loss and decrease in harvest index.

Several studies have suggested that leaf reflectance of corn (Blackmer et al., 1994; Zhao et al., 2003), cotton (Zhao et al., 2005a), and sorghum (Zhao et al., 2005b) at wavelengths of 550 and 710 nm are most sensitive to N supply. Our study indicates that cotton canopy reflectance response to the N treatments depends on both growth stage and wavelength (data not shown). The low N treatment (56N) mainly increased canopy reflectance centered 550 and 710 nm during squaring and fruiting. This is in agreement with the results of leaf reflectance response to N

rate in other crops (Blackmer et al., 1994; Zhao et al., 2003, 2005b). Therefore, the reflectances at 550 and 710 ( $\pm 5$ ) nm may be used to detect plant N deficiencies of most field crops. Seasonal patterns of RVI and NDVI (Fig. 3) during cotton growth and development in the present study are consistent with those in corn (Gitelson et al., 2003) and winter wheat (Serrano et al., 2000). In general, RVI and NDVI are two major indicators of ground coverage of vegetation canopy and of canopy greenness (Li et al., 2001). Nitrogen deficiency reduces LAI or canopy size (Fernandez et al., 1996; Zhao and Oosterhuis, 2000; Reddy et al., 2004) and leaf photosynthetic pigment contents of cotton (Wood et al., 1992; Zhao and Oosterhuis, 2000; Zhao et al., 2005a), probably resulting in low values of RVI, NDVI, and other indices. Among the tested indices, RVI and  $dR/d\lambda$  could be used to most clearly distinguish the N treatments.

Similar to previous reports in corn (Gitelson, 2004) and winter wheat (Serrano et al., 2000), the relationships of cotton LAI and ABM to most tested indices followed hyperbolic trends. Both RVI and NDVI did not increase with further increases in LAI and ABM (*i.e.* saturation) when cotton LAI or ABM reached a certain level during growth (see Fig. 4). Some studies have reported that hyperspectral reflectance indices of MTVI2 and MCARI2 (Haboudane et al., 2004) or broadband WDRVI (Gitelson, 2004) can avoid the saturation at high LAI. In the present study, WDRVI, MCARI, MCARI/OSAVI, MTVI2, MCARI2 and  $dR/d\lambda$  had less saturation with LAI compared to NDVI, but any of them did not significantly improve relationships between the reflectance and LAI or ABM in cotton (Fig. 4). We found that when logarithm transformation of LAI and ABM was used, relationships of  $\log(\text{LAI})$  and  $\log(\text{ABM})$  with most of tested reflectance indices became linear (Fig. 4). Among the reflectance indices, WDRVI and OSAVI had the greatest  $r^2$  with  $\log(\text{LAI})$  and  $\log(\text{ABM})$ . Therefore, using log values of LAI and ABM can improve the projected relationships of LAI and ABM with the reflectance indices.

Correlation between lint yield and canopy reflectance indices depended both on reflectance index and on cotton growth stage at which the reflectance measurements were made (Fig. 5). To improve crop yield prediction from remote sensing of canopy reflectance, therefore, selection of proper measuring date is important. It is evident that early to peak bloom stage is the best time for measuring canopy reflectance in order to accurately estimate lint yield. When compared with RVI or NDVI, any modified reflectance indices (*i.e.* EVI, WDRVI, MCARI, MCARI, OSAVI, MTVI2, and MCARI2) did not significantly improve the relationships between canopy reflectance and relative lint yield in most measuring dates in the present study (Fig. 5). This is in agreement with Zarco-Tejada et al. (2005). Our results of weak correlation between lint yield and the reflectance indices at early growth stage agree with Bronson et al. (2005). The poor relationship between relative lint yield and most reflectance indices, calculated from reflectance data collected at early growth stages in the present study, was probably associated with a low LAI. Aparicio et al. (2000) similarly reported a weak correlation between grain yield and RVI or NDVI in durum wheat when LAI was low. In soybean, Ma et al. (2001) documented a correlation between seed yield and NDVI measured at the R2 to R5

reproductive stages. Moges et al. (2004) also found a close relationship between grain yield and NDVI in winter wheat at Feekes growth stage 5. However, the relationships of soybean seed and wheat grain yields with NDVI, reported by Ma et al. (2001) and Moges et al. (2004), followed a power or exponential function rather than the linear function. Our results indicated that cotton relative lint yield was associated with RVI, NDVI, EVI, and WDRVI at any growth stage between pre-flowering and first boll opening, and the greatest  $r^2$  value was achieved around the early flower stage in both years ( $r^2 = 0.56\text{--}0.89$ ,  $P < 0.001$ ). Therefore, these canopy reflectance indices and linear algorithms developed around early flower stage may be used to predict cotton yield. Additionally, validation of the regression models (Fig. 6) of four major reflectance indices (*i.e.* RVI, NDVI, EVI, and WDRVI) suggested that the performance of the RVI regression model was better than that of the other models for cotton yield prediction.

## 5. Conclusions

Nitrogen fertilizer rate and year significantly influenced cotton lint yield, but yield did not increase linearly with increasing amounts of N application rate. During early growth, cotton canopy reflectance indices, RVI, NDVI, EVI, WDRVI, OSAVI, MTVI2, MCARI2, and  $dR/d\lambda$  increased rapidly with progression of plant age and achieved maximum values around early flower stage. Symptoms of N deficiency stress were especially evident in 2002 for plants in the 0N treatment, which had significantly lower LAI and ABM during flowering and boll development than N-fertilized plants. Correlations of lint yield with reflectance indices depended on plant growth stage at which canopy reflectance data were collected. Treatment differences in lint yield were linearly correlated with changes in most reflectance indices measured between early square and boll development stage. The coefficients (intercept and slope) and  $r^2$  values of the linear regression models between lint yield and the reflectance indices indicated that early flower stage (70–75 DAS) was the best time to measure canopy reflectance in cotton for prediction of relative lint yield. Most tested canopy reflectance indices may be used to determine canopy structure of high-yielding, irrigated cotton at early flower stage, to assess lint yield, and to help growers make field management decisions during the growing season.

## Acknowledgements

This research was funded in part by the National Aeronautical and Space Administration through the GeoResources Institute at Mississippi State University. We express our appreciation to D. Brand, K. Gourley and A.R. Mohammed for technical support. Contribution from the Department of Plant and Soil Sciences, Mississippi State University, Mississippi Agricultural and Forestry Experiment Station, paper no. J-10751.

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