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DETERMINATION OF COTTON NITROGEN STATUS WITH A HAND-HELD CHLOROPHYLL METER[1]

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ABSTRACT: The ability of a hand-held chlorophyll meter (SPAD-502 Chlorophyll Meter[3], Minolta Camera Co., Ltd., Japan) to determine the N status of cotton (*Gossypium hirsutum* L.) was studied at field sites in Alabama and Missouri. Meter readings on the uppermost fully-expanded leaf were compared to leaf-blade N and petiole NO_3-N at first square, first bloom and midbloom as to their seed cotton yield predictive capability. Nitrogen was applied at rates of 0, 45, 90, 135, 180 and 225 kg ha^{-1} to establish a range of cotton chlorophyll levels, tissue N concentrations, and seed cotton yields. A typical curvilinear cotton yield response to N fertilizer was observed in Alabama experiments.

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Because of adverse weather conditions, cotton yield in Missouri experiments did not respond to N. Chlorophyll meter readings were significantly correlated to leaf-blade N concentration at all three stages of growth for all experiments. In Alabama, chlorophyll meter readings compared favorably to leaf-blade N and petiole NO_3-N with respect to their seed cotton yield predictive capability at all three stages of growth. It appears that hand-held chlorophyll meters would be as reliable as leaf-blade N and petiole NO_3-N for predicting supplemental N fertilization requirements of cotton. However, more research will be required prior to use of chlorophyll meter readings for routine cotton-N recommendation purposes.

INTRODUCTION

Producers, consultants and researchers have long sought a quick, reliable method to diagnose cotton N status during the growing season. The goal has been to develop a means of tailoring N fertilizer programs to specific conditions under which a cotton crop is grown. Too little N early in the growing season limits vegetative growth and boll set, while excess N promotes rank vegetative growth, boll rot and delayed maturity. In addition to economic consequences, excess N fertilization can cause contamination of ground and surface waters.

Methods currently used for predicting cotton N requirements include soil and tissue testing. Soil NO_3-N tests prior to cotton planting have been successful in predicting cotton N requirements in the western U.S. (Gardner and Tucker, 1967), but have been less effective in the humid Southeast (Lutrick et al., 1986). Tissue tests, including petiole NO_3-N and leaf-blade N, have become popular means of monitoring cotton N status in many production areas. Cotton petiole NO_3-N testing has shown good N requirement predictive capability in Arkansas, Georgia and Florida (Lutrick et al., 1986), Oklahoma (Baker et al., 1972) and Texas (Sunderman et al., 1979), but has been less successful in Alabama (Touchton et al., 1981), Mississippi (Jenkins et al., 1982) and Tennessee (Howard and Hoskinson, 1986). Leaf-blade N analyses are less affected by climate and seasonal changes than petiole NO_3-N tests (Sabbe and Zelinski, 1990). This allows the producer to sample the crop in a less rigorous manner than would be possible with petiole NO_3-N testing, because of less change in leaf-blade N concentration as the growing season progresses.

Hand-held chlorophyll meters may offer a good corollary to leaf-blade N analyses, and, thus, aid in prediction of N requirement for cotton. One such meter, the SPAD-502 Chlorophyll Meter[3] (Minolta Camera Co., Ltd., Japan) has demonstrated effectiveness in predicting the N status of rice (*Oryza sativa* L.) (Takebe et al., 1990; Takebe and Yoneyama, 1989; Kitigawa et al., 1987; Turner and Jund, 1991) and corn (*Zea mays* L.) (Wood et al., 1991; Schepers et al., 1990). The principle of measurement and operation of this device has been described elsewhere (Inada, 1963; Wood et al., 1991). Briefly, the SPAD-502 meter detects the difference in leaf-blade light attenuation at 430 and 750 nm, and displays a numerical SPAD (Soil Plant Analysis Development) unit, ranging from 0 to 80.

Chlorophyll meter readings are instantaneous and involve no tissue collection. Therefore, if cotton N status can be determined with these meters, costs associated with sample collection and laboratory analyses could be reduced substantially. In addition, chlorophyll meters could enable cotton producers to respond to N deficiencies in a more timely fashion than previously possible.

The objectives of our study were to 1) determine the feasibility of using hand-held chlorophyll meters for evaluation of cotton N status, and to 2) compare leaf chlorophyll measurements with leaf-blade N and petiole NO_3-N analyses as to their seed cotton yield predictive capability.

METHODS

The ability of a hand-held chlorophyll meter to predict cotton N status was tested in Alabama and Missouri during 1991. In each state, one irrigated and one non-irrigated experiment were planned. However, in Alabama, because of the high amount of growing-season rainfall (Fig. 1), no supplemental irrigation was applied to the planned irrigated experiment. Alabama experiments were on a

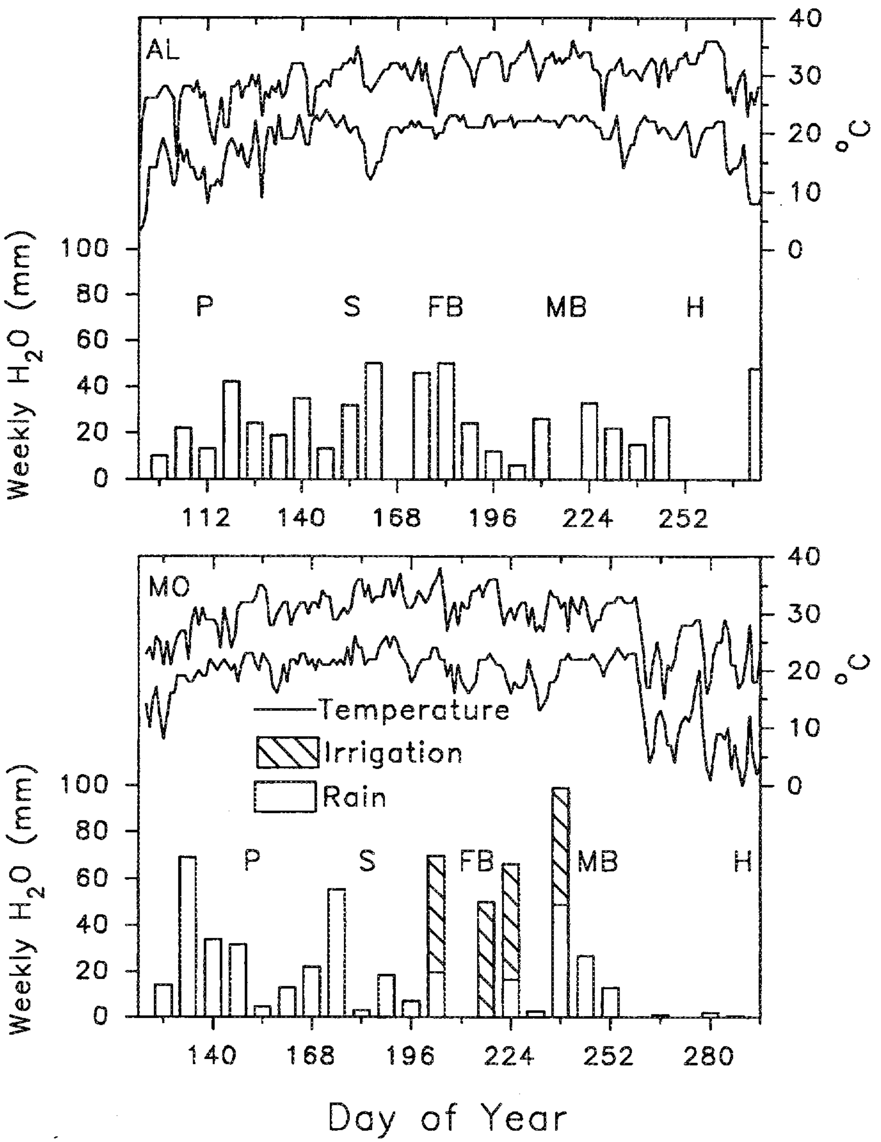

Fig. 1. Growing Season Rainfall, Irrigation and Maximum and Minimum Temperatures for Alabama and Missouri Experiments. Planting Date = P, First Square = S, First Bloom = FB, Mid-Bloom = MB, and Harvest = H.

Norfolk sandy loam (fine-loamy, siliceous thermic Typic Kandiudults) at the E.V. Smith Research Center of the Alabama Agricultural Experiment Station near Shorter, AL. Missouri experiments were on a Tiptonville fine sandy loam (fine-silty, mixed, thermic Typic Argiudoll) at the Delta Research Center of the Missouri Agricultural Experiment Station at Portageville, MO.

All four experiments were managed as a conventionally tilled cotton production system with the goal of optimum lint yields. Except for irrigation, cultural practices were identical for irrigated and non-irrigated experiments in a particular state. In Alabama, soil pH (0 to 15 cm) was 6.2 (1:1 soil:H_2O), and P, K, S, Mg and Zn were preplant, broadcast applied (according to Auburn University soil test recommendations) to all plots and incorporated. Cotton (Deltapine 50[3]) was planted at a rate of 19 seed m^{-1} with a John Deere Flex 71 planter[3] on 22 April. In Missouri, soil pH (0 to 15 cm) was 5.4 (1:1 soil:0.01*N* $CaCl_2$). Because of high soil test levels, fertilization with nutrients other than N was not necessary. Cotton (Stoneville 506[3]) was planted at a rate of 16 seed m^{-1} with a John Deere Max-Emerge 7300 planter[3] on 31 May. Row spacing was 97 cm in both states. Cotton was planted on raised (10 cm height) and flat seedbeds in Missouri and Alabama, respectively.

The irrigated experiment in Missouri received 200 mm of supplemental moisture via four 50 mm irrigation events (Fig. 1). Water was delivered to cotton in that experiment with a surface furrow irrigation system.

Chemical weed control in Alabama consisted of preemergence applications of norflurazan (4-chloro-5-(methylamino)-2-(3-trifluoromethyl)phenyl)-3(2*H*)-pyridazinone) at 1.4 kg a.i. ha^{-1} and fluometuron (*N,N*-dimethyl-*N'*-[3-(trifluoromethyl)phenyl]urea) at 1.1 kg a.i. ha^{-1}. In Missouri, weeds were controlled via preplant incorporation of trifluralin (2,6-dinitro-*N,N*-dipropyl-4(trifluoromethyl)benzenamine) at 0.8 kg a.i. ha^{-1} and norflurazin at 1.4 kg a.i. ha^{-1}. Additional weed control in all experiments was achieved by cultivation as needed.

All experiments were scouted weekly for insect pests, and appropriate chemicals were applied as necessary. No unusual insect infestations were noted in any of the experiments.

Experimental designs were randomized complete blocks of four replications, and were identical in Alabama and Missouri. In all experiments, fertilizer N treatments were broadcast by hand upon stand establishment to effect a range of cotton chlorophyll levels, tissue N concentrations, and seed cotton yields. Nitrogen rates were 0, 45, 90, 135, 180 and 225 kg N ha^{-1}. The N source in Alabama was NH_4NO_3, while N was applied as urea in Missouri. Individual plot size was 8.1 X 7.6 m.

In Alabama experiments, cotton began producing squares (first square) 42 days after planting (DAP). First square occurred 31 DAP in Missouri experiments. Blooms began opening (first bloom) 60 DAP in Alabama experiments. In Missouri experiments, first bloom occurred 58 DAP. Midbloom occurred 100 and 90 DAP in Alabama and Missouri, respectively. At first square, first flower and midbloom, chlorophyll measurements (10 $plot^{-1}$) were made on the uppermost fully expanded leaf with a Minolta SPAD-502 chlorophyll meter[3]. On the same day as chlorophyll measurements, 30 of the uppermost fully expanded leaves in each plot were collected. Petioles were severed from leaves for NO_3-N analysis. Leaf blades were retained for total N analysis. Petioles and leaf blades were dried at 60°C and ground to pass a 0.5 mm sieve. Leaf-blade tissue and petioles from Missouri experiments, and petioles from Alabama experiments, were analyzed by the University of Arkansas Soil Testing Laboratory, Marianna, AR. Leaf blades from Alabama experiments were analyzed for total N in the Department of Agronomy and Soils at Auburn University with a LECO CHN-600 analyzer[3].

In Alabama, the two center rows of each plot were harvested for seed cotton yield with a spindle cotton picker on 16 September (147 DAP). Similarly, seed cotton in Missouri was harvested on 24 October (147 DAP).

Data were analyzed via regression procedures with the SAS package[3] (SAS Institute, 1988). The proposed adequate linear model included linear and quadratic terms. Dummy variables (Draper and Smith, 1981) for irrigation and irrigation by other independent variable interactions were tested to determine deviations between irrigated and non-irrigated experiments in Missouri. Stepwise elimination of nonsignificant independent variables was utilized, and terms were eliminated from the models if nonsignificant at the α = 0.10 level. Because no supplemental irrigation was applied to the irrigated experiment in Alabama (Fig. 1), we made no attempt to separate irrigation effects for the Alabama experiments.

RESULTS AND DISCUSSION

Abundant, well distributed precipitation during the growing season in Alabama experiments (Fig. 1) promoted high seed cotton yields and response to N fertilizer. A typical curvilinear seed cotton yield response to applied N was observed (Fig. 2). Solving the first derivative of the yield response curve gave a maximum agronomic yield of 3.54 Mg seed cotton ha^{-1} at 207 kg N ha^{-1}. Based on a ginning fraction of 0.38, a lint price of \$1.43 kg^{-1} and a N cost of \$0.6 kg^{-1}, a maximum economic yield of 3.46 Mg seed cotton ha^{-1} was obtained at 191 kg N ha^{-1}.

Growing season weather also played a role in determining cotton response to N in Missouri experiments. Although yields were high (Fig. 2), ranging from 2.5 to 3.4 Mg seed cotton ha^{-1}, N applications had no significant effect on yield. Planting date in Missouri experiments was delayed due to heavy rainfall during late April and early May (Fig. 1), which compressed the growing season. Where N was applied, cotton maturity was delayed; this has been observed in other studies (Maples and Frizzell, 1985). Consequently, low temperatures during September and October (Fig. 1) prevented bolls from opening, resulting in the lack of response to N fertilizer. Seed cotton yield was approximately 0.5 Mg ha^{-1} greater in the irrigated than in the non-irrigated experiment across all N rates.

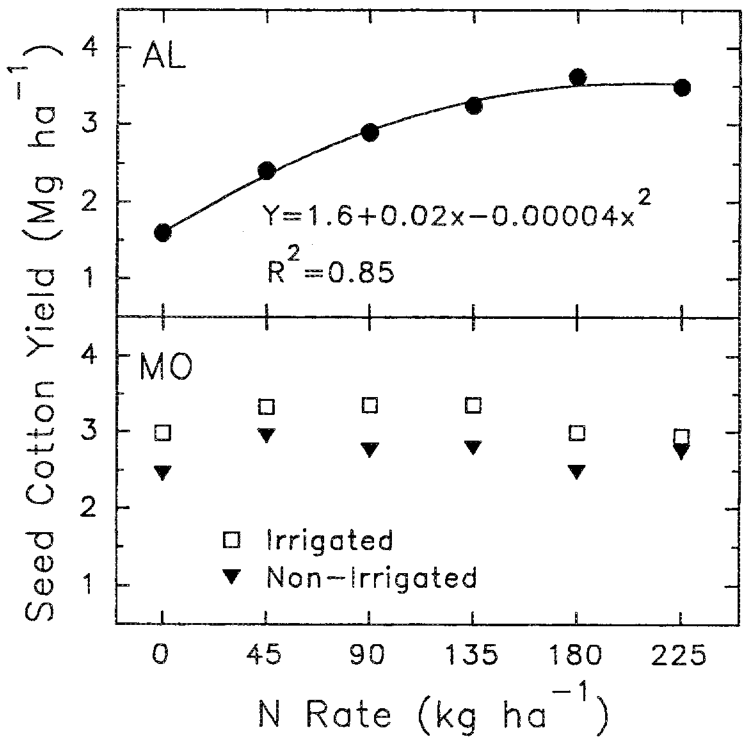

Fig. 2. Seed Cotton Yield Response to N Fertilizer in Alabama and Missouri Experiments.

The relationship between chlorophyll meter readings on the first fully expanded leaf and tissue N concentrations of the same plant part for Alabama and Missouri experiments is shown in Fig. 3. Chlorophyll meter readings were highly correlated to tissue N concentrations at all three stages of growth. A narrower range of leaf-blade N concentrations was observed in Missouri than in Alabama experiments (Fig. 3). The smaller leaf-blade N range could have been due to a higher N soil fertility regime under Missouri study sites as evidenced by the higher lower limit of tissue N at midbloom in Missouri vs. Alabama (Fig. 3).

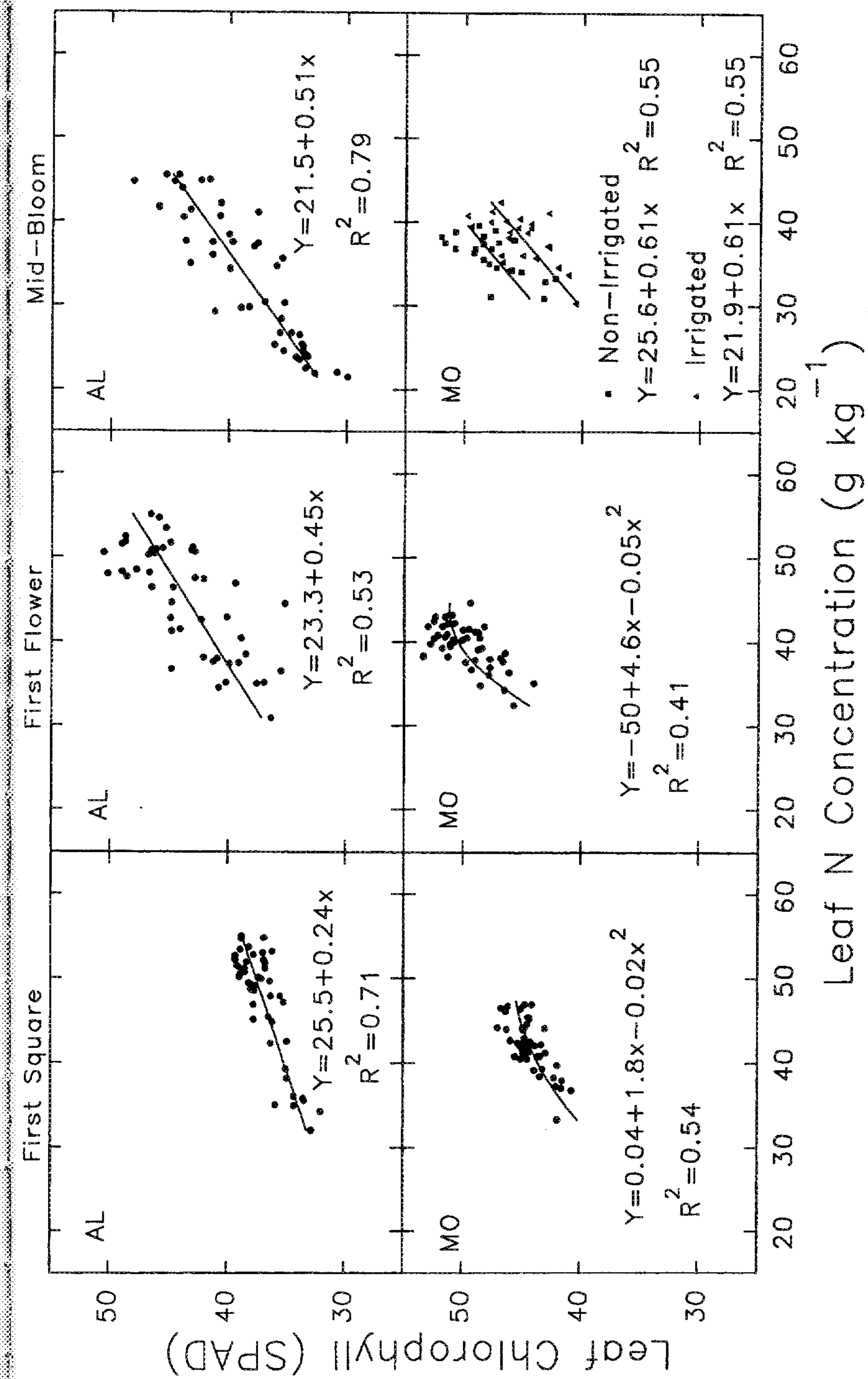

Fig. 3. Relationship Between Leaf Blade Total N Concentrations and Chlorophyll Meter Readings at First Square, First Bloom, and Mid-Bloom in Alabama and Missouri Experiments.

The regressions for Missouri experiments at first square and first bloom were quadratic, which indicates a buildup of leaf non-chlorophyll N at those stages of growth. Chlorophyll meter readings were lower in the irrigated than in the non-irrigated experiment at midbloom, due to a greater non-chlorphyll N:chlorophyll N ratio under irrigation.

Relationships between seed cotton yield and leaf-blade N concentration at first square, first bloom and midbloom for Alabama experiments are shown in Fig. 4. Leaf-blade tissue N concentration was a good predictor of seed cotton yield at all three stages of growth. Nitrogen concentration of first fully-expanded leaf blades declined during the growing season. This decline indicated that, as the season progressed, dry matter accumulation rates were greater than N uptake and that N was being shunted to reproductive structures (Oosterhuis et al., 1983). Maximum economic yield (3.46 Mg seed cotton ha^{-1}) was obtained with 58, 54 and 40 g N kg^{-1} leaf-blade tissue at first square, first bloom, and midbloom, respectively. Economic response to N fertilizer would be expected below these leaf-blade N concentrations. First square and midbloom leaf-blade N concentrations corresponding to maximum economic yield are, however, well above sufficiency levels reported by workers in Arkansas (30 to 43 g N kg^{-1}) and Georgia (35 to 45 g N kg^{-1}) (Sabbe and Zelinski, 1990), and probably reflect the nature of the 1991 growing season in Alabama.

As expected from the high correlation between leaf-blade tissue N concentrations and chlorophyll meter readings (Fig. 3), chlorophyll meter readings in Alabama were a good predictor of seed cotton yield (Fig. 4). Leaf chlorophyll readings at maximum economic yield were 39, 49 and 47 at first square, first bloom and midbloom, respectively. Chlorophyll meter readings were not as highly correlated to seed cotton yields as leaf-blade N. However, it appears that this tool may offer an alternative to chemical tissue tests, particularly when convenience is considered. The capability of leaf chlorophyll readings to predict seed cotton yields at first square is especially promising, because supplemental N could easily be applied at that stage of growth. More research

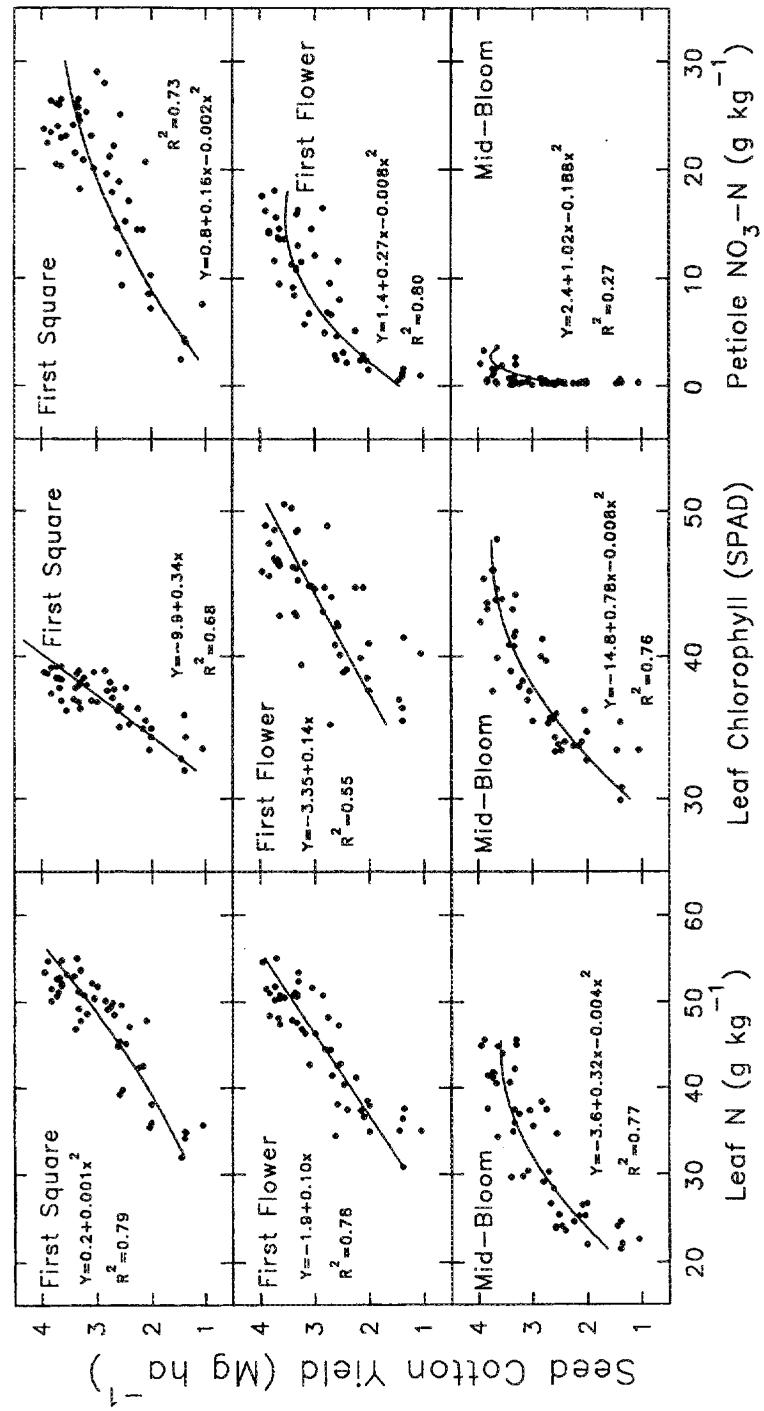

Fig. 4. Relationships Between Seed Cotton Yield and Leaf-Blade N, Chlorophyll Meter Readings and Petiole NO_3-N at First Square, First Bloom and Mid-Bloom in Alabama Experiments.

will, however, be required to calibrate this tool for prediction of supplemental N fertilization requirements for cotton. Since cotton N status varies with variety and cultural practices (Sabbe and Zelinski, 1990), studies that address chlorophyll meter readings in relation to varietal differences and management are needed.

Petiole NO_3-N tests were included in the experiments as a comparison to leaf chlorophyll readings, because they are used extensively in the U.S. for monitoring cotton N status. Petiole NO_3-N levels typically decline as the growing season progresses, so that sufficiency levels vary with growth stage. Tucker (1963), working in Arizona, recommended sufficiency levels of 15, 12, 7, and 4 g NO_3-N kg^{-1} petiole tissue at first square, first bloom, first boll, and first open boll, respectively. In California, MacKenzie et al. (1963) set sufficiency levels of 16, 8 and 2 g NO_3-N kg^{-1} petiole tissue at the early, mid-, and late-bloom stages of growth, respectively. Petiole NO_3-N data from our Alabama experiments followed previously published patterns; petiole NO_3-N concentrations corresponding to maximum economic yield were 24, 12, and 1.5 g NO_3-N kg^{-1} at the first square, first bloom, and midbloom stages, respectively (Fig. 4). At first square and first bloom, petiole NO_3-N concentrations were a better predictor of seed cotton yields than chlorophyll meter readings. However, at midbloom, chlorophyll meter readings had superior seed cotton yield predictive capability when compared to petiole NO_3-N concentrations.

As previously mentioned, adverse weather conditions disallowed seed cotton yield responses to N fertilizer in Missouri experiments. Therefore, no significant correlations between seed cotton yield and leaf-blade N concentration, chlorophyll meter reading or petiole NO_3-N concentration were observed in Missouri experiments (data not shown).

CONCLUSIONS

Chlorophyll meter readings compared favorably to standard leaf-blade and petiole NO_3-N tests for evaluation of cotton N status. This technology deserves further attention. Additional research should be conducted prior to use of chlorophyll meters as a tool for cotton N requirement prediction.

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GROWTH AND NITROGEN (N_2) FIXATION RESPONSE OF ARROWLEAF CLOVER TO MINERAL NITROGEN AND 2(N-MORPHOLINO)-ETHANESULFONIC ACID AT LOW pH[1]

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ABSTRACT: Evaluation of legume response to acidic conditions can be difficult when using nutrient solutions because of fluctuations in solution pH. The organic buffer 2(N-morpholino)-ethanesulfonic acid (MES) has been used for stabilizing pH in nutrient solution studies. We evaluated the effectiveness of MES (5.0 m*M*) to stabilize solution culture at pH 5.5 with and without mineral N (0 or 1.0 m*M* NH_4NO_3) and its influence on growth and N_2 fixation of arrowleaf clover (*Trifolium vesiculosum* Savi). The buffer maintained pH stability ± 0.1 pH units in the presence or absence of mineral N. In the absence of mineral N, the quantity of N_2 fixed by plants grown with MES was not significantly different from that fixed by plants grown without MES. However, with mineral N, N_2 fixation was reduced 37% with addition of MES. Tissue analysis indicated a small increase in Ca and Mg concentration for plants grown with MES. Caution should be exercised in the use of MES in studies of N_2-fixing legumes when mineral N is included.

INTRODUCTION

Nutrient solution systems aid in evaluating nutritional and environmental influences on growth and N_2 fixation activity of legumes. However, pH can be difficult to maintain because N_2 fixation alters solution pH (1). Methods for

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