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Using a Chlorophyll Meter to Determine the Chlorophyll Concentration, Nitrogen Concentration, and Visual Quality of St. Augustinegrass

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Abstract. **Because high rates of nitrogen fertility are necessary for producing high-quality turfgrasses, quick, reliable methods of determining the N status of turfgrasses would be valuable management tools. The objective of this study was to evaluate the capacity of a hand-held chlorophyll meter (SPAD-502) to provide a relative index of chlorophyll concentrations, N concentrations, and visual quality in St. Augustinegrass [*Stenotaphrum secondatum* (Walt.) Kuntze]. Two experiments were conducted in a greenhouse in 1998 to evaluate the utility of SPAD readings. Established pots of 'Floratam' were subjected to weekly foliar Fe treatments at Fe rates of 0 and 0.17 kg·ha^{-1} for 4 weeks. Six weekly nitrogen fertilizer treatments were applied in the form of ammonium sulfate at N rates of 0, 5.75, 11.5, 17.25, and 23 kg·ha^{-1} for 4 weeks. Greenhouse SPAD readings were not affected by Fe treatment, but N treatments resulted in differences in SPAD readings, visual quality, and chlorophyll concentrations. The readings were positively correlated with chlorophyll concentrations (r^2 = 0.79), visual ratings (r^2 = 0.74), and total Kjeldahl nitrogen (TKN) (r^2 = 0.71). Readings taken from field-grown 'Floratam', 'Floratine', and 'Floralawn' St. Augustinegrass were poorly correlated (r^2 < 0.63) with chlorophyll concentrations and TKN. Unless future techniques improve dependability of the SPAD meter under field conditions for measuring chlorophyll and N concentration of a stand of turfgrass, the usefulness of such readings for the management of St. Augustinegrass seems limited.**

The Soil-Plant Analyses Development (SPAD) unit of Minolta Camera Co. has developed the SPAD-502 chlorophyll meter (Minolta Camera Co., Japan), a hand-held, self-calibrating, convenient, and nondestructive lightweight device used to calculate the amount of chlorophyll present in plant leaves (Minolta, 1989; Yadava, 1985). This meter records optical density measurements at two wavelengths, converts them into digital signals, and then into a SPAD value (Minolta, 1989). Such values have been positively correlated (r^2 = 0.93) with destructive chlorophyll measurements in rice (*Oryza sativa* L.), wheat (*Triticum aestivum* L.), and soybean [*Glycine max* (L.) Merr.] (Monje and Bugbee, 1992), and with extractable chlorophyll for 11 food crop species (r^2 > 0.90) (Marquad and Tipton, 1987). Strong relationships between leaf N concentration and SPAD values were found in apple (*Malus* ×*domestica* Borkh.), but varied within the growing season (Neilsen et al., 1995). Following adjustments based on leaf weight to account for development stages, SPAD values were correlated (r^2 = 0.97) with leaf N concentration in corn (*Zea mays* L.) (Chapman and Barreto, 1997). Chlorophyll meter readings, along with other simple measurements such as soil nitrate tests, have the potential to quickly and easily predict the N requirements for winter wheat (Reeves et al., 1993). These studies suggest that proper interpretation of SPAD values could be a useful tool in developing N fertilizer programs.

No research correlating tissue chlorophyll concentrations with N concentration in warm-season turfgrasses has been reported. Chlorophyll contents have been positively correlated (r^2 = 0.91) with visual turf ratings (Madison and Anderson, 1963). A linear relationship (r^2 > 0.90) was indicated between chlorophyll meter readings and extractable chlorophyll in forage-type fescue (*Festuca arundinaceae* Schreb.) (Kantety et al., 1993). One limitation for the use of the SPAD-502 chlorophyll meter on turfgrass is the narrow leaf blade common to most turfgrasses. The SPAD measurement area is 2 × 3 mm (Minolta, 1989), which makes measurements of narrow-leafed turfgrasses such as bermudagrass [*Cynodon dactylon* (L.) Pers.] difficult. A wider (4–10 mm) leaf blade makes St. Augustinegrass a good candidate for SPAD measurements. Further research is needed to determine the potential for using such measurements for managing N fertility in St. Augustinegrass. The objective of this study was to evaluate the ability of the SPAD-502 Chlorophyll Meter to measure the relative leaf chlorophyll concentrations, N concentrations, and visual quality in St. Augustinegrass.

Materials and Methods

Two greenhouse studies were conducted in Spring and Summer 1998 to evaluate the utility of a hand-held chlorophyll meter (model SPAD-502; Minolta Corp., Ramsey, N.J.) to assess the chlorophyll status and to correlate it with N status and visual quality in St. Augustinegrass. 'Floratam' St. Augustinegrass sod was washed thoroughly to remove soil and cut into 12-cm-diameter plugs; these were placed on sand in 16-cm-diameter plastic pots and topdressed with sand. Pots were placed in a glasshouse maintained at an average day temperature of 31 °C and an average night temperature of 29 °C under mist irrigation for 7 weeks until established. To aid in establishment, pots were fertilized with 24 kg·ha^{-1} N as $(NH_4)_2SO_4$ 6 weeks prior to starting treatments. Pots were arranged in a split-plot design with two Fe fertilization treatments as main plots and five N fertilization treatments as subplots in four replications. Two foliar Fe fertilizer treatments were applied weekly for 4 weeks at Fe rates of 0 and 0.17 kg·ha^{-1} and five N fertilizer treatments were applied at N rates of 0, 5.75, 11.5, 17.25, and 23 kg·ha^{-1} as $(NH_4)_2SO_4$ over the same period. SPAD readings and visual color ratings were taken every 2 weeks beginning 6 weeks after fertilizer treatments were started for a total of three sampling dates. At each sampling date, pots were rated visually for color on a scale of 1–10 with 1 representing a dead stand and 10 representing highest quality. One to two mature leaves were collected per pot and five SPAD measurements were taken per leaf and averaged. Collected leaves were then prepared for chlorophyll extraction using dimethyl sulfoxide (Hiscox and Israelstam, 1979). Chlorophyll concentrations were determined by subjecting spectrophotometer (Spectronic 21; Milton Roy Co., Rochester, N.Y.) absorbance values to an equation (Arnon, 1949). Chlorophyll concentration and SPAD data were collected every 2 weeks for a total of three samplings. After the third SPAD sampling, additional data were taken to assess N status. One SPAD reading from each of ten randomly selected mature leaves was taken and averaged for each pot. Each pot was clipped to a height of 3 cm using scissors and an aluminum catch pan to collect clippings. Clippings were dried in a forced-air oven at 75 °C for 48 h and

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evaluated for total Kjeldahl N (TKN) using wet chemistry methods (Jones, 1991). Corresponding visual ratings, SPAD measurements, chlorophyll concentrations, and N concentrations were compared using the SAS General Linear Model and PROC CORR procedures (SAS Inst., 1987).

Additional data were taken on 11 Aug. 1998 to assess SPAD meter readings under field conditions. Samples were taken from 'Floratam', 'Floratine', and 'Floralawn' St. Augustinegrass foundation blocks grown on Arredondo fine sand (loamy, siliceous, Typic Quartzipsamment) at the G.C. Horn Memorial Turfgrass Field Laboratory in Gainesville, Fla. Each cultivar block was divided into sixteen 3 × 3 m plots for sampling. One or two mature leaves were collected per plot and five SPAD measurements were taken per leaf and averaged. Collected leaves were then prepared for chlorophyll extraction and chlorophyll concentrations were determined. An additional twenty SPAD measurements were taken at random within each plot and averaged. Each plot was then harvested by mowing at a height of 7.5 cm using a commercial walk-behind rotary mower and a custom chute and plexiglass catcher attachment. Clippings were dried in a forced-air oven at 75 °C for 48 h, and TKN was determined by wet chemistry methods (Jones, 1991). Chlorophyll and N concentrations, and SPAD measurements were compared using the SAS General Linear Model and PROC CORR procedures (SAS Inst., 1987).

Results and Discussion

Greenhouse SPAD readings were not affected by Fe treatment, but N treatments affected SPAD readings, visual quality, chlorophyll concentrations, and N concentrations, which was their intended purpose (Table 1). The range of N fertility treatments was applied for the purpose of establishing a range of chlorophyll concentrations, N concentrations, and levels of visual quality for correlation. For this reason, differences among N treatments were not analyzed further. SPAD readings were positively correlated ($r^2 = 0.79$) with chlorophyll concentrations (Fig. 1) and with visual rating ($r^2 = 0.74$), which was expected since visual ratings were based on color (Fig. 2). Since chlorophyll contents have previously been correlated with visual turfgrass ratings (Madison and Anderson, 1963), these data suggest that SPAD readings may be a more objective means of measuring visual quality based on color in St. Augustinegrass. A positive correlation ($r^2 = 0.71$) was found between SPAD reading and TKN concentration (Fig. 3), as expected based on the SPAD–chlorophyll correlation in this study and on previous SPAD–N relationships in other crops (Chapman and Barreto, 1997; Neilsen et al., 1995). These results suggest that SPAD readings may be a rapid method of assessing the chlorophyll status and that they are correlated with chlorophyll concentration, visual quality, and N status of St. Augustinegrass grown under controlled conditions.

Table 1. Mean squares from analysis of variance of SPAD readings, chlorophyll concentrations, and visual ratings on greenhouse-grown 'Floratam' St. Augustinegrass.

		Mean square			
Source of variation	df	SPAD reading	Chlorophyll concentration	Visual rating	Total Kjeldahl N
Experiment, (Exp)	1	140.72*	0.66**	1.01	792.93**
Error A	3	14.91	0.26	0.88	138.96
Fe treatment, (F)	1	19.7	0.47	2.81	138.96
Error B	3	12.1	0.11	1.20	33.37
N treatment, (N)	4	136.51**	0.45**	39.16**	6419.31**
Exp × N	4	23.52*	0.05	4.54*	38.87
Exp × F	1	45.45*	0.52*	0.11	50.73
F × N	4	2.79	0.31	0.78	61.47
Exp × F × N	4	2.45	0.01	0.27	46.04
Error C	51	8.4	0.05	0.52	33.53

*, **Significant at $P < 0.05$ and 0.01, respectively.

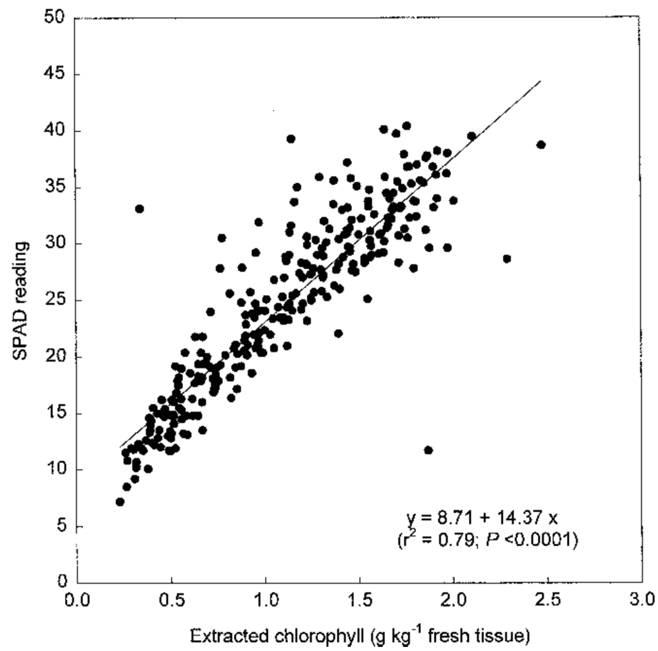

Fig. 1. Relationship between SPAD reading and extracted chlorophyll for greenhouse-grown 'Floratam' St. Augustinegrass.

Correlations in the field study were not as strong as those in the greenhouse. There were positive correlations between SPAD readings and chlorophyll concentrations for all three cultivars used in this study (Fig. 4). Positive correlations were also found between SPAD readings and TKN concentrations (Fig. 5) although r^2 values were lower than those for SPAD–chlorophyll correlations. This may be due to sampling method, because SPAD readings were taken from the actual leaves used for chlorophyll extraction while representative SPAD readings were taken across an entire plot before collecting clippings for N determination. Field ranges for TKN and chlorophyll concentrations were also narrow; no treatments were applied to create a broad range, which may partially explain low r^2 values. Although linear relationships were found, r^2 values were much lower than those found in the greenhouse experiment.

Further research is needed to assess the usefulness of SPAD readings for the management of St. Augustinegrass. While greenhouse results were promising, field results were not as good. This may be due to variability within an area of established turfgrass and difficulty in taking truly representative samples. More positive results found in previous studies of other crops may be related to uniformity of the development stages of the crop studied (Chapman and Barreto, 1997; Reeves et al., 1993). A stand of turfgrass is comprised of many individual plants at many stages of development, and relating results taken from a few individual leaves to a larger area of established turf is difficult. Another possible shortcoming of the SPAD meter is leaf size. Although St. Augustinegrass has a wide leaf relative to other turfgrasses, sampled leaves must invariably include a portion of the midvein, which was a source of variation in a previous chlorophyll meter study (Monje and Bugbee, 1992). Unless future techniques or instrumentation improve the dependability of the SPAD meter for measuring chlorophyll and N concentration of a stand of turfgrass

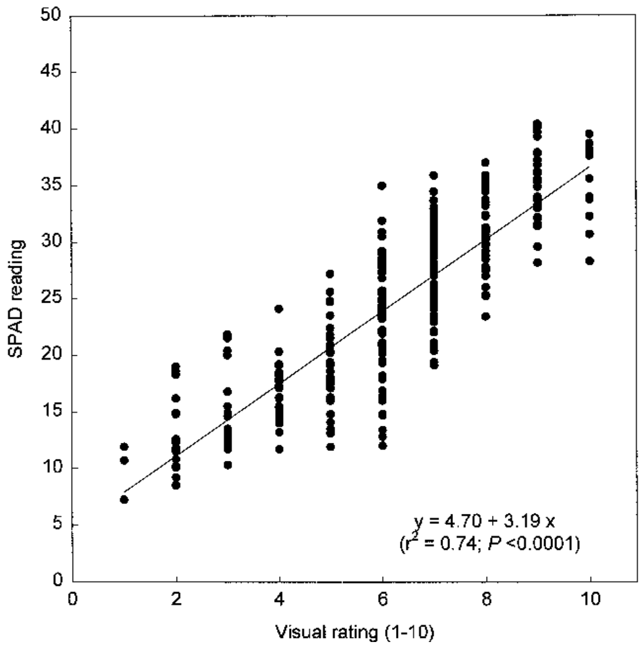

Fig. 2. Relationship between SPAD reading and visual rating for greenhouse-grown 'Floratam' St. Augustinegrass.

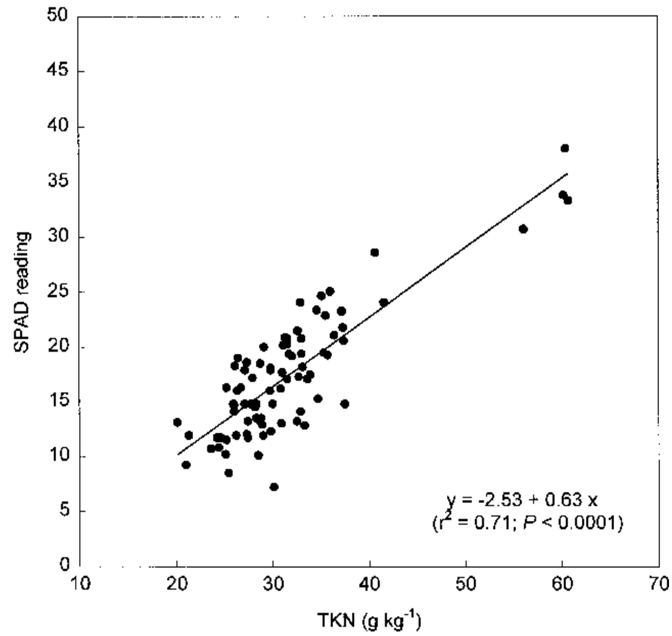

Fig. 3. Relationship between SPAD reading and total Kjeldahl nitrogen (TKN) for greenhouse-grown 'Floratam' St. Augustinegrass.

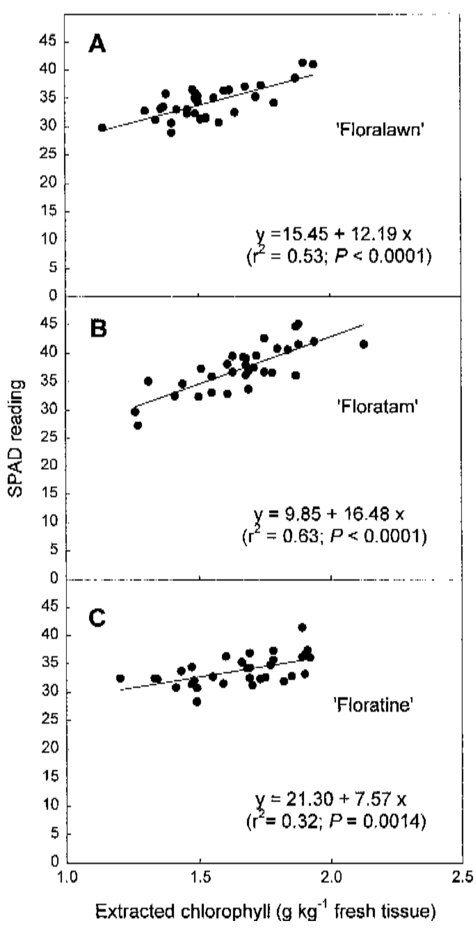

Fig. 4. Relationship between SPAD reading and extracted chlorophyll for field-grown (**A**) 'Floralawn', (**B**) 'Floratam', and (**C**) 'Floratine' St. Augustinegrass.

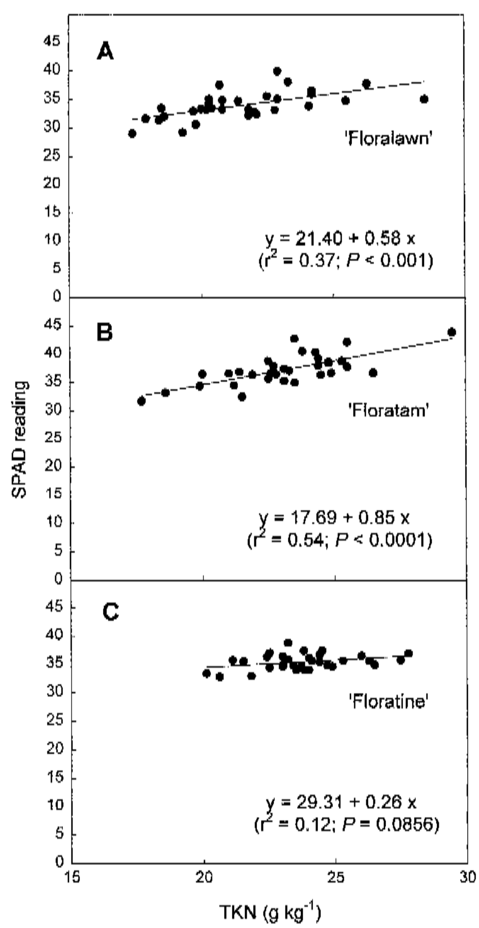

Fig. 5. Relationship between SPAD reading and total Kjeldahl nitrogen (TKN) for field-grown (**A**) 'Floralawn', (**B**) 'Floratam', and (**C**) 'Floratine' St. Augustinegrass.

under field conditions, the usefulness of SPAD readings for the N management of St. Augustinegrass seems limited.

In summary, SPAD-502 chlorophyll meter readings were affected by applications of N but not of Fe. SPAD readings correlated well with chlorophyll concentrations, visual quality, and TKN in St. Augustinegrass grown under greenhouse conditions, but were a poor indicator of chlorophyll and N concentrations of St. Augustinegrasses grown in the field.

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