Detection of nitrogen and phosphorus nutrient status in bermudagrass using spectral radiance

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ABSTRACT

Nitrogen (N) and phosphorus (P) are two of the most limiting nutrients for crop production. Because of this, continued interest focuses on improving N- and P-use efficiency. Spectral radiance measurements were evaluated to identify optimum wavelengths for dual detection of N and P status in bermudagrass (*Cynodon dactylon* L.). A factorial arrangement of treatments (0, 112, 224, and 336 kg N ha^{-1} and 0, 29, and 58 kg P ha^{-1}) was applied to an established bermudagrass pasture for further study using a randomized complete block design. A wide range of spectral radiance measurements (276-831 nm) was obtained from each plot using a PSD1000 Ocean Optics fiber optic spectrometer. The resulting spectra were partitioned into 10-nm bands.

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Added indices were generated to test for correlation of N and P content with spectral radiance. The 435-nm band (430-440 nm) was found to be independent of N and P treatment, and as a covariate, significantly decreased residual error. Using 435 nm as a covariate, it was found that biomass, N uptake, P uptake, and N concentration could be predicted using 695/405. No index reliably predicted bermudagrass forage P concentration. Spectral radiance has the potential to be used for predicting N and P nutrient status, but further work is needed to document response in different environments.

INTRODUCTION

Recent work has documented micro-variability in soil test N and P, thus enhancing the potential use of sensor-based-variable-rate-technology (s-VRT) for fertilizer application. Optimizing fertilizer application using s-VRT may reduce N and P fertilizer input costs as well as surface and ground water contamination from over fertilization.

Light reflectance can be used to detect the nutrient element status of plants because plants have strong absorption of light by chlorophyll and near infrared reflectance (NIR) (Thomas and Oerther, 1972). In addition, organic compounds have unique absorption properties due to vibration of molecular bonds (Morra et al., 1991). However, several factors affect the reflectance such as nonuniformity of incident solar radiation, plant structure, leaf area, background reflectivity (tillage), diseases, physiological stress (Knipling, 1970) and leaf thickness (Wooley, 1971). Nutrient status affects crop performance and visual symptoms, therefore, the effect of fertilizer on plant growth may be detected using spectral radiance.

There are several existing indices that have been used to predict nutrient status. In corn, N status was estimated using spectral radiance at 550 nm (Blackmer et al., 1994) and NIR/red (Walburg et al., 1982). In winter wheat, NDVI (normalized difference vegetation index = NIR-red / NIR+red) was used to predict N uptake and biomass (Stone et al., 1996). Milton et al. (1991) found that P deficiencies resulted in higher reflectance in the green and yellow portions of the electromagnetic spectrum. Little research has addressed the use of spectral radiance on detecting plant P deficiencies. The objective of this research was to identify indices for the dual detection of N and P status in bermudagrass.

MATERIALS AND METHODS

One study was conducted on an established bermudagrass (*Cynodon dactylon* L.) pasture near Burneyville, Oklahoma, on a Minco fine sandy loam (coarse-silty, mixed thermic Udic Haplustoll). Initial soil test characteristics are reported in Table 1. An N rate x P rate factorial arrangement of treatments (0, 112, 224, and 336 kg N ha^{-1} with 0, 29, and 58 kg P ha^{-1}) was evaluated in a randomized complete block experimental design with three replications. Nitrogen and P fertilizers were both

TABLE 1.   Initial surface (0-15 cm) soil test characteristics, Burneyville, OK, 1996.

Characteristics	Extractant	Unit	Value
pH	1:1 soil:H_2O		5.68
Organic Carbon[ω]	Dry Combustion	$g\,kg^{-1}$	9.564
Total Nitrogen[ω]	Dry Combustion	$g\,kg^{-1}$	0.872
NH_4-N[Π]	2 M KCl	$mg\,kg^{-1}$	13.1
NO_3-N[Π]	2 M KCl	$mg\,kg^{-1}$	6.4
P[Ψ]	Mehlich 3	$mg\,kg^{-1}$	23.8
K[Ψ]	Mehlich 3	$mg\,kg^{-1}$	163.5

[ω]Schepers et al. (1989).
[Π]Lachat Instruments (1989).
[Ψ]Mehlich (1984).

applied on April 18, 1996, and August 9, 1996. The plots were 3.1x9.1 m with 6.1 m alleys. Spectral readings and forage yields were collected on May 29, 1996 (the first harvest after the first fertilization), June 27, 1996 (the second harvest after the first fertilization), August 9, 1996 (the third harvest after the first fertilization), and September 13, 1996 (the first harvest after the second fertilization).

Spectral data were collected within each plot using a PSD1000 portable dual spectrometer manufactured by Ocean Optics, Inc., from two overlapping bandwidths of 276-831 nm and 650-1,100 nm. The fiber optic spectrometer which has 200 μm diameter and no slit has spectral resolution as low as 5 nm. The bifurcated fiber was lifted with an hemispherical lucite™ lens which increased its angle of acceptance to 34°. The lens was held at 1.5 m and the area sensed was 0.8 m^2 plot^{-1}.

All spectral readings were partitioned into 10 nm bandwidths. In addition to these spectral bands collected from each reading, the spectral indices NDVI (Normalized Difference Vegetation Index) and other combinations of single indices were generated. Total N in forage was determined using a Carlo-Erba (Milan, Italy) NA 1500 Dry Combustion Analyzer (Schepers et al., 1989). Total P was determined using a nitric-perchloric acid (NHO_3-$HClO_4$) digest and concentration determined as per a modified method developed by Murphy and Riley (1962). Nitrogen and P uptake was calculated by multiplying N or P concentration by biomass. Individual wavelengths and combinations of wavelengths (or ratios) were used to predict agronomic responses such as N and P concentration, N and P uptake, and biomass. Statistical analyses included regression, analysis of variance, and covariance. All were performed using procedures defined in SAS (SAS Institute, 1988). Since the wavelengths and indices were not consistent over time, analysis of covariance was performed for spectrometer readings. The selection criteria for covariates

TABLE 2. Analysis of variance for total forage N, N uptake, total P concentration, P uptake, May 29 and June 27, Burneyville, OK, 1996.

Source of variation	Last fertilization: April 18, 1996 Harvest: May 29, 1996					Last fertilization: April 18, 1996 Harvest: June 27, 1996				
	Biomass	N Conc.	N uptake	P Conc.	P Uptake	Biomass	N Conc.	N uptake	P Conc.	P uptake
	Mean squares									
Rep	ns	ns	ns	ns	ns	ns	**	ns	***	ns
N rate	***	***	***	***	***	ns	***	**	***	*
P rate	ns	ns	ns	**	ns	ns	ns	ns	ns	ns
NxP	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Residual	207217	1.5	80.92	15434	0.755	1256671	3.4	711.1	65790	9.8
CV, %	18	5.7	16.0	6	17.1	41	7.1	37.7	9	39.6
Contrasts:										
N linear	***	***	***	***	***	ns	***	***	***	**
N quadratic	***	**	***	**	***	ns	ns	ns	**	ns
P linear	ns	ns	ns	***	ns	ns	*	ns	ns	ns
P quadratic	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns

N, kg·ha^{-1}	kg·ha^{-1}	g·kg^{-1}	kg·ha^{-1}	mg·kg^{-1}	kg·ha^{-1}	kg·ha^{-1}	g·kg^{-1}	kg·ha^{-1}	mg·kg^{-1}	kg·ha^{-1}
0	1667	14.4	24.6	1597	2.7	2151	18.3	38.8	2265	4.9
112	2738	20.3	55.4	1902	5.2	2897	22.5	65.4	2713	7.8
224	3043	23.8	71.4	2137	6.4	3132	29.1	86.6	3314	9.8
336	2781	26.6	72.8	2131	5.9	2821	32.9	91.8	3224	9.2
SED	215	0.6	4.2	59	0.4	528	0.9	12.6	121	1.5
P, kg·ha^{-1}										
0	2481	21.2	53.9	1823	4.6	2652	26.4	69.3	2829	7.5
29	2633	21.3	59.0	1967	5.3	2728	26.0	72.2	2972	8.2
58	2558	21.2	55.2	2035	5.3	2870	24.7	70.5	2837	8.1
SED	186	0.5	3.7	51	0.4	458	0.7	10.9	105	1.3

*,**,***Significant at the 0.05, 0.01, and 0.001 probability levels, respectively.

ns=not significant.

SED=standard error of the difference between two equally replicated means.

TABLE 3. Analysis of variance for total forage N, N uptake, total P concentration, P uptake, August 9 and September 13, Burneyville, OK, 1996.

Source of variation	Last fertilization: April 18, 1996 Harvest: August 9, 1996					Last fertilization: August 9, 1996 Harvest: September 13, 1996				
	Biomass	N Conc.	N uptake	P Conc.	P uptake	Biomass	N Conc.	N uptake	P Conc.	P uptake
					-----Mean squares-----					
Rep	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
N rate	ns	***	***	***	**	***	***	***	***	***
P rate	ns	ns	ns	ns	ns	ns	**	ns	ns	ns
NxP	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Residual	898846	1.9	402.3	119479	15.5	1913863	1.2	946.4	105319	17.0
CV, %	18	7.4	20.5	9	19.5	24	5.1	24.2	9	19.6
Contrasts:										
N linear	ns	***	***	***	**	**	***	***	***	***
N quadratic	ns	ns	ns	ns	ns	**	*	**	***	**
P linear	ns	ns	ns	ns	ns	ns	*	ns	ns	ns
P quadratic	ns	ns	ns	ns	ns	ns	*	ns	ns	ns

N, kg·ha⁻¹	kg·ha⁻¹	g·kg⁻¹	kg·ha⁻¹	mg·kg⁻¹	kg·ha⁻¹	kg·ha⁻¹	g·kg⁻¹	kg·ha⁻¹	mg·kg⁻¹	kg·ha⁻¹
0	4765	14.1	67.7	3285	16.0	3703	16.9	62.5	3095	11.5
112	5583	17.8	97.9	3662	20.4	6932	20.7	143.1	3252	22.4
224	4885	20.5	98.0	4093	19.9	6268	23.2	142.9	4026	24.9
336	5762	22.4	126.7	4238	24.6	6386	25.2	159.8	4059	25.3
SED	447	0.7	9.5	163	1.9	652	0.5	14.5	153	1.9
P, kg·ha⁻¹										
0	4987	19.1	93.9	3837	19.4	5504	22.3	124.6	3574	19.6
29	5273	18.4	95.9	3763	20.2	5912	20.8	126.6	3588	21.7
58	5487	18.6	103.1	3858	21.1	6049	21.3	129.9	3661	21.9
SED	387	0.6	8.2	141	1.6	565	0.4	12.6	132	1.7

*,**,***Significant at the 0.05, 0.01, and 0.001 probability levels, respectively.

ns=not significant.

SED=standard error of the difference between two equally replicated means.

TABLE 4. Analysis of variance for selected indices from various readings using 435 nm as a covariate for readings on May 29 and June 27, Burneyville, OK, 1996.

Source of variation	df	725/535	695/405	NDVI	PNSI	805/695
May 29, 1996		----------------------------------Mean squares----------------------------------				
Rep	2	***	ns	ns	ns	ns
N rate	3	**	***	***	***	***
P rate	2	*	ns	ns	ns	ns
NxP	6	ns	ns	ns	ns	ns
w435	1	***	ns	ns	ns	ns
Contrasts:						
N linear	1	***	***	***	***	***
N quadratic	1	ns	***	**	**	*
P linear	1	*	ns	ns	ns	ns
P quadratic	1	ns	ns	ns	ns	ns
Error	21	0.003	0.003	0.000	0.003	0.019
CV, %		2.0	2.5	2.7	2.7	4.1
N rate, kg ha^{-1}		----------------------------------Means----------------------------------				
0		2.765	2.427	0.513	1.949	3.116
112		2.871	2.254	0.549	1.822	3.438
224		2.860	2.242	0.549	1.823	3.440
336		2.900	2.229	0.555	1.801	3.500
SED		0.027	0.027	0.007	0.024	0.066
P rate, kg ha^{-1}						
0		2.810	2.297	0.534	1.872	3.299
29		2.865	2.293	0.545	1.839	3.408
58		2.872	2.275	0.546	1.836	3.413
SED		0.023	0.024	0.006	0.021	0.057

June 27, 1996		-----------------Mean squares-----------------				
Rep	2	ns	ns	ns	ns	ns
N rate	3	ns	*	ns	ns	ns
P rate	2	ns	ns	ns	ns	ns
NxP	6	ns	ns	ns	ns	ns
w435	1	***	ns	ns	ns	*
Contrasts:						
N linear	1	ns	*	*	ns	*
N quadratic	1	ns	*	ns	ns	ns
P linear	1	*	ns	ns	ns	ns
P quadratic	1	ns	ns	ns	ns	ns
Error	21	0.005	0.014	0.002	0.031	0.113
CV, %		3.2	4.9	8.1	9.1	10.6
N rate, kg·ha^{-1}		-----------------Means-----------------				
0		2.892	2.584	0.487	2.080	2.942
112		3.033	2.386	0.523	1.923	3.215
224		3.080	2.439	0.528	1.909	3.265
336		3.057	2.417	0.530	1.906	3.295
SED		0.035	0.057	0.028	0.084	0.159
P rate, kg·ha^{-1}						
0		2.301	2.461	0.501	2.026	3.065
29		2.328	2.456	0.520	1.939	3.194
58		2.380	2.452	0.529	1.899	3.279
SED		0.030	0.049	0.024	0.72	0.137

*,**,***Significant at the 0.05, 0.01, and 0.001 probability levels, respectively.

ns=not significant.

SED=standard error of the difference between two equally replicated means.

TABLE 5.   Analysis of variance for selected indices from various readings using 435 nm as a covariate for readings August 9 and September 13, Burneyville, OK, 1996.

Source of variation	df	725/535	695/405	NDVI	PNSI	805/695
August 9, 1996				-----Mean squares-----		
Rep	2	ns	ns	ns	ns	ns
N rate	3	ns	*	ns	ns	ns
P rate	2	ns	ns	ns	ns	ns
NxP	6	ns	ns	ns	ns	ns
w435	1	*	***	**	**	**
Contrasts:						
N linear	1	ns	**	ns	ns	ns
N quadratic	1	ns	ns	ns	ns	ns
P linear	1	ns	ns	ns	ns	ns
P quadratic	1	ns	ns	ns	ns	ns
Error	21	0.011	0.003	0.000	0.345	0.001
CV, %		6.5	1.1	6.7	6.8	1.6
N rate, kg·ha^{-1}				-----Means-----		
0		1.628	4.573	0.119	8.548	1.270
112		1.627	4.569	0.120	8.347	1.273
224		1.582	4.540	0.116	8.708	1.263
336		1.576	4.496	0.115	8.806	1.259
SED		0.050	0.025	0.004	0.277	0.009
P rate, kg·ha^{-1}						
0		1.592	4.551	0.116	8.683	1.263
29		1.579	4.535	0.117	8.603	1.264
58		1.639	4.548	0.119	8.521	1.271
SED		0.042	0.021	0.003	0.240	0.008

September 13, 1996		Mean squares				
Rep	2	ns	ns	ns	ns	ns
N rate	3	***	***	***	***	***
P rate	2	ns	ns	ns	ns	ns
NxP	6	ns	ns	ns	ns	ns
w435	1	***	***	***	***	***
Contrasts:						
N linear	1	**	***	***	***	***
N quadratic	1	***	***	***	***	***
P linear	1	ns	ns	ns	ns	ns
P quadratic	1	ns	ns	ns	ns	ns
Error	21	0.001	0.008	0.000	0.002	0.089
CV, %		1.0	4.1	2.8	3.2	6.2
N rate, kg·ha^{-1}		Means				
0		2.576	2.600	0.585	1.720	3.901
112		2.630	2.128	0.669	1.503	5.164
224		2.631	2.141	0.669	1.499	5.111
336		2.615	2.187	0.660	1.517	4.931
SED		0.129	0.014	0.003	0.023	0.141
P rate, kg·ha^{-1}						
0		2.627	2.277	0.641	1.574	4.709
29		2.603	2.279	0.645	1.561	4.734
58		2.609	2.236	0.652	1.545	4.887
SED		0.011	0.012	0.002	0.020	0.122

*,**,***Significant at the 0.05, 0.01, and 0.001 probability levels, respectively.
ns=not significant.
SED=standard error of the difference between two equally replicated means.

$$y = -33.304x + 97.474$$
$$R^2 = 0.5226$$
$$Pr>F=0.0001$$
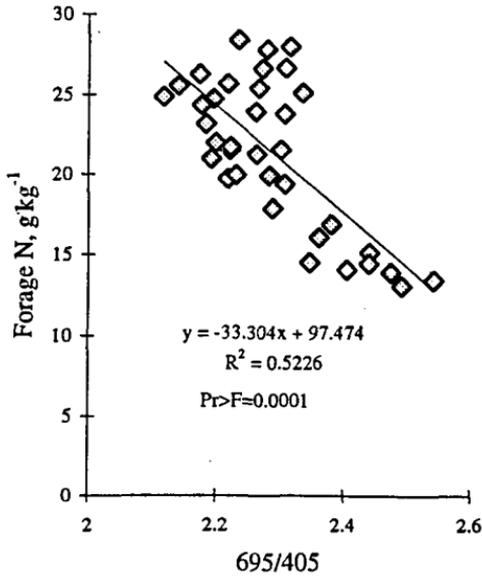
FIGURE 1.   Correlation between bermudagrass N concentration and 695/405 on May 29, 1996.

$$y = -7.4086x + 38.094$$
$$R^2 = 0.3083$$
$$Pr>F = 0.0005$$
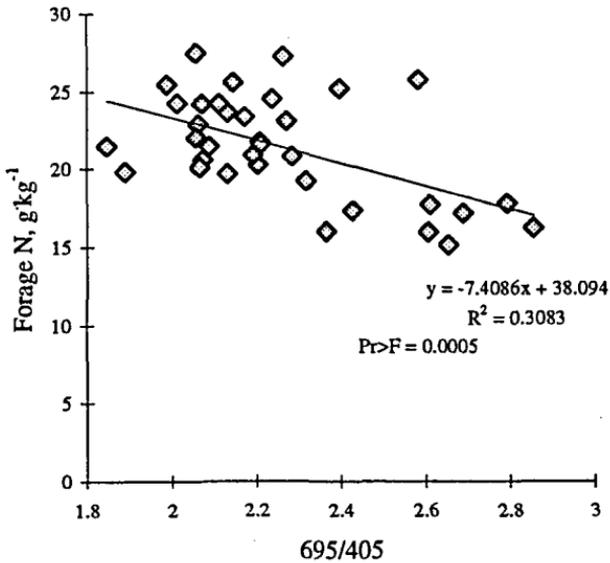
FIGURE 2.   Correlation between bermudagrass N concentration and 695/405 on September 13, 1996.
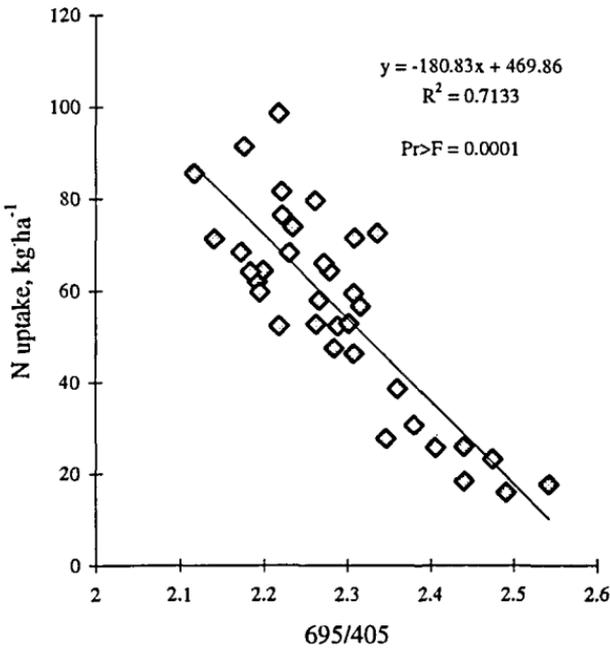
FIGURE 3. Correlation between bermudagrass N uptake and 695/405 on May 29, 1996.

included no significant effect of treatment and no correlation with dependent variables. Bandwidths which adhered to these criteria were 375, 395, 435, 445, 455, 465, 475, 485, 495, 785, 795, and 805 nm. The indices used to predict agronomic responses were chosen based on similarity in AOV.

RESULTS AND DISCUSSION

Agronomic Responses

Analysis of variance models with factorial effects of N, P, and N x P are reported in Tables 2 and 3 for biomass, N, N uptake, P and P uptake for the three dates where comprehensive data were collected. Similar to the first fertilization, no significant interaction of N and P was detected for the second fertilization (Tables 2 and 3) thus allowing direct interpretation of main effects of N and P independently. Forty-one days following the first fertilization, a significant quadratic response to N fertilization was found for dry biomass, N concentration, N uptake, P concentration, and P uptake. This result suggests that N was limiting response since increases
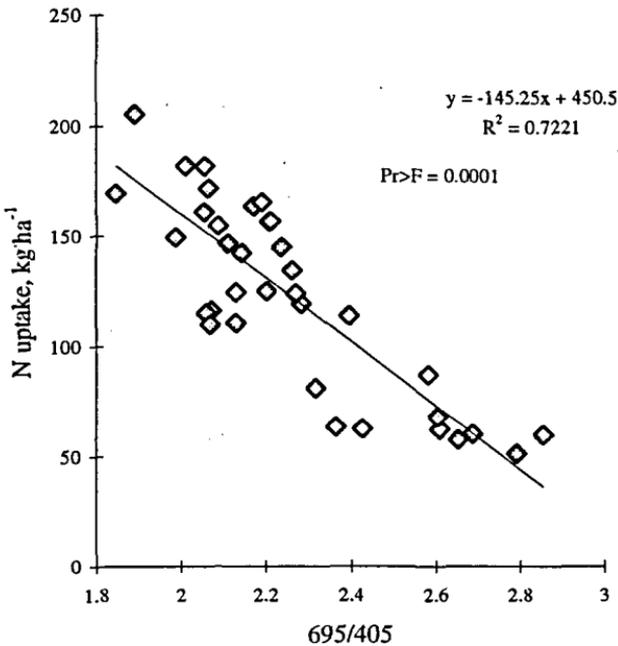
FIGURE 4.   Correlation between bermudagrass N uptake and 695/405 on September 13, 1996.

with N rate were shown for biomass, N concentration and N uptake, P concentration and P uptake. It was interesting to find that N rate influenced P concentration in bermudagrass tissue. As N rate increased, P concentration increased (Tables 2 and 3). Following 71 and 113 days from the first fertilization, a linear response to N rate was observed for N concentration, and N and P uptake. Tissue P concentration responded linearly to N applied 41 days following the first fertilization. A linear trend for increased N to increase biomass, N concentration, N uptake, P concentration and P uptake by the second fertilization was also observed (Table 3).

Spectrometer Readings

If significant differences in measured biological variables were found, we expected reflectance changes to be significant as well. However, no one index was consistently related to measured plant response over time. It was thought that the consistency of one index over multiple sampling dates could be increased using P
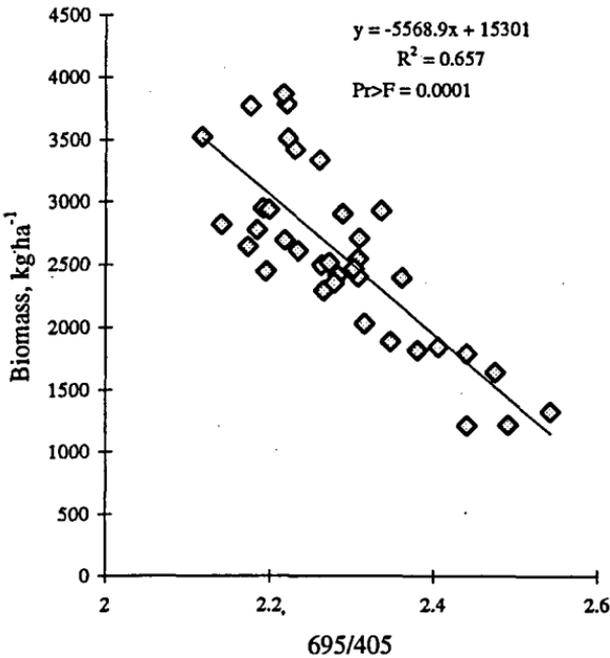
FIGURE 5.   Correlation between bermudagrass biomass and 695/405 on May 29, 1996.

covariance. The 435 nm band (430-440 nm) was found to be independent of N and treatment, and as a covariate, significantly decreased residual error (Tables 4 and 5). This was consistent over several cuttings, but results did vary with time in terms of the percentage error accounted for by the 435 nm covariate.

The indices which behaved similarly to observed differences in N concentration over time (means and significance of AOV) were 695/405 and NDVI. The effect of N rate was highly significant for N concentration as well as 695/405 and NDVI. The relationship between 695/405 and forage N concentration for the May 29, 1996 and September 13, 1996 dates is reported in Figures 1 and 2, respectively. Although both indices were not highly correlated with forage N, they were significant (Probability of greater F value from the model, P>F).

Similar to results for N concentration, 695/405 was highly correlated with N uptake (Figures 3 and 4). It was important to find a consistent, positive relationship with N uptake even when values changed significantly with time (100 kg N ha⁻¹ versus 200 kg N ha⁻¹ from May 29 to September 13, 1996).

No consistent index or 10 nm band was correlated with P concentration at any sampling date. However, similar to N uptake, the index that best predicted P uptake was 695/405. This index was obviously providing good prediction of biomass
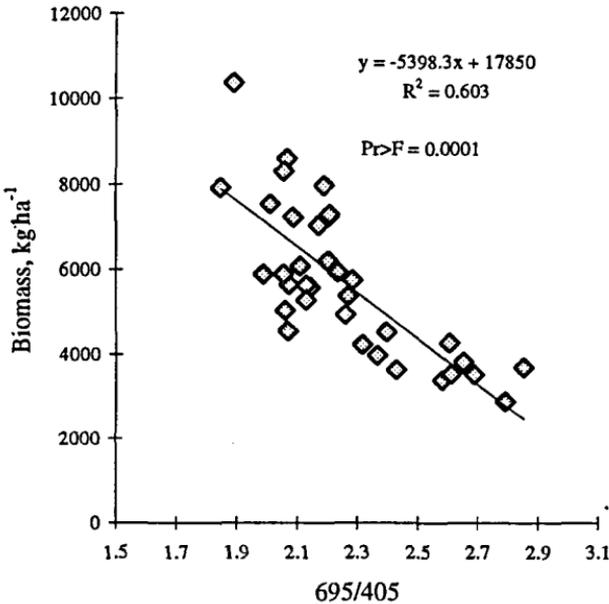
FIGURE 6.   Correlation between bermudagrass biomass and 695/405 on September 13, 1996.

since similar response in N and P uptake was observed. Correlation between 695/405 and P uptake was good for both May 29 and September 13 harvest dates. It is important to note that several indices were positively correlated with biomass (NDVI and 695/405). NDVI has been commonly used to predict biomass which was consistent with what is reported here. Correlation between biomass and 695/405 is presented in Figures 5 and 6 for May 29, 1996 and September 13, 1996; and biomass with NDVI in Figures 7 and 8, respectively. Using a linear model, correlation with dry biomass was consistently better using 695/405 when compared to NDVI.

CONCLUSIONS

The 435 nm band (430-440 nm) was found to be independent of N and P treatment, and as a covariate, significantly decreased residual error. Using 435 nm as a covariate, it was found that N uptake, P uptake, and N concentration could be predicted using 695/405; whereas, biomass was best predicted using NDVI. However, no index reliably predicted bermudagrass forage P concentration. Spectral radiance has the potential to be used for predicting N and P nutrient status, but further work is needed to document response in different environments.
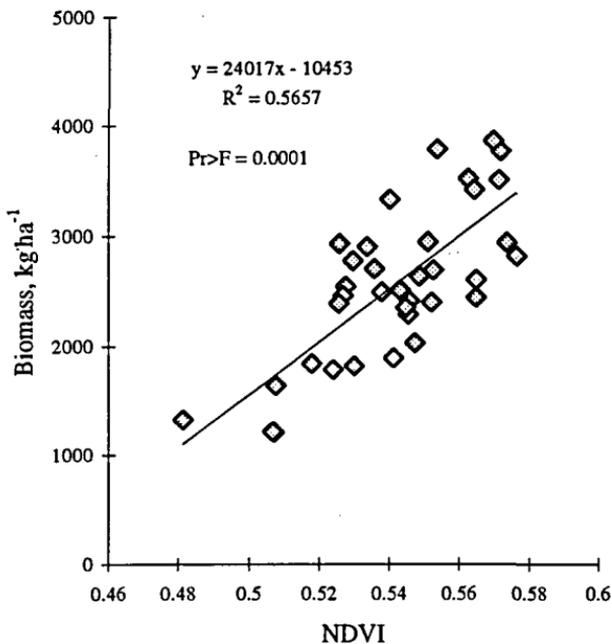
FIGURE 7. Correlation between bermudagrass biomass and NDVI on May 29, 1996.
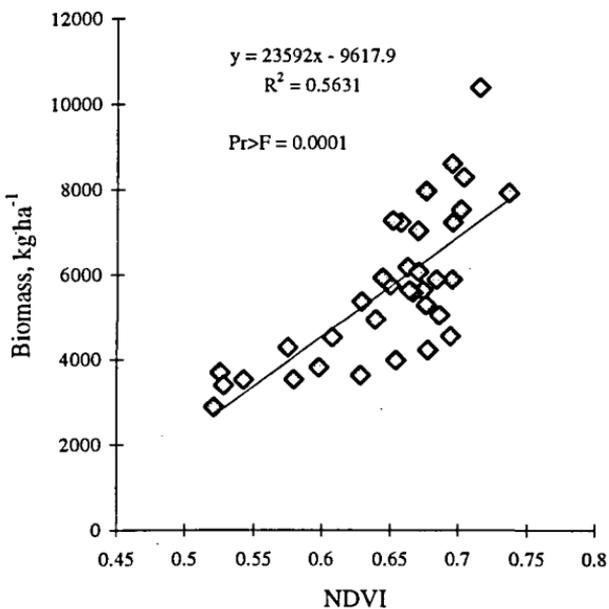
FIGURE 8. Correlation between bermudagrass biomass and NDVI on September 13, 1996.

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