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Estimating crude protein concentration of a grass sward using spectral measurements

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Introduction

While measurement of forage mass (FM) is fundamental to evaluating sward productivity, its forage quality determines the value for grazing. Optical sensing devices have varying potential for estimating forage characteristics. Instruments such as the Greenseeker[®], which measures normalized difference vegetation index (NDVI) have been related to various characteristics, but are confounded with canopy traits (Biewer *et al.* 2009). Simultaneous readings of NDVI and FM were used to separate their contributions to sward crude protein CP using multiple regression (Moyer *et al.* 2010).

Relationships in swards of tall fescue (*Lolium arundinacium* Schreb.) were poorer than in other species, so further spectroscopic study in that species may indicate how estimates of forage quality, specifically CP concentration, might be improved.

Methods

Instrumentation

Spectral reflectance data were obtained with an Analytical Spectral Devices (ASD) FieldSpec 3 portable spectrometer (ASD Inc., Boulder, CO, USA). It measures wavelengths from 0.3 to 2.5 μm , with a sampling interval of 1.4 nm at 350 – 1000 nm, and 2.0 nm at 1000 – 2500 nm. A Greenseeker RT200 sensor (Trimble, Ukiah, CA) ahead of a mobile rising plate unit was also used to measure sward NDVI.

Data collection

Tall fescue plots 12 \times 1.5 m were established at the Kansas State University North Agronomy farm, Manhattan, KS USA. Fertilizer N was applied in fall, 2011 at increments from zero to 224 kg/ha in replicated blocks to stimulate differences in spring forage CP.

On 4 April, 2012, ~ 500 g of forage was hand-clipped near one end of each plot, sealed, and refrigerated (4°C) for ~24 hr. Spectral readings were taken in the laboratory with the ASD FieldSpec 3 from two types of fescue samples in: (1) fresh leaf surfaces, and (2) dried, ground

forage. Six readings of fresh leaves, or 10 readings per dry sample were taken from petri dishes using a Muglight in an otherwise dark environment. Dried forage had been at 60°C for five days and ground to pass a 0.5 mm sieve, then used for N analysis.

The ASD FieldSpec 3 was also used to obtain spectral readings in the field. Ten canopy readings per plot were taken on 6 April, a clear day, within an hour of solar noon. Height above the canopy was ~1 m, providing readings from a diameter of about 0.4 m.

On 18 April, the rising plate unit with the Greenseeker[®] RT200 sensor was used to estimate FM and NDVI. A plot area 4.6 \times 0.36 m was harvested, weighed, and a subsample collected, dried, and ground for dry weight and N analysis. Reduced N concentration was determined by H₂SO₄-H₂O₂ digestion and subsequent colorimetric assay, or multiplied by 6.25 for expression as CP.

Data analysis

The ASD spectral readings were converted to reflectance data and analyzed using the GRAMS/AI 8.0 software (Thermo Electron Corp., Salem, New Hampshire, USA), as in El Masry *et al.* (2007). Partial least square (PLS) regression coupled with a cross-validation algorithm was performed on the datasets to select significant PLS factors with N as the dependent variable.

Results

ASD FieldSpec 3

Mean spectral reflectance curves from field plots and lab samples were analyzed with the corresponding N (thus CP) concentrations, resulting in coefficient of determination (R^2) values of 72%, 67%, and 65% for field-obtained, lab-assayed fresh leaves, and lab-assayed ground samples, respectively. Principle-factor analysis indicated that the largest amount of variation in N concentrations was explained by 2, or at most 3 spectral factors. Figure 1 shows the principal factor loadings to occur in the 1421-1600 nm and 2200-2400 nm spectral regions. Those two spectral regions in the near-infrared (NIR) range explained the greatest amount of variation in N (thus CP) within the tall fescue plots.

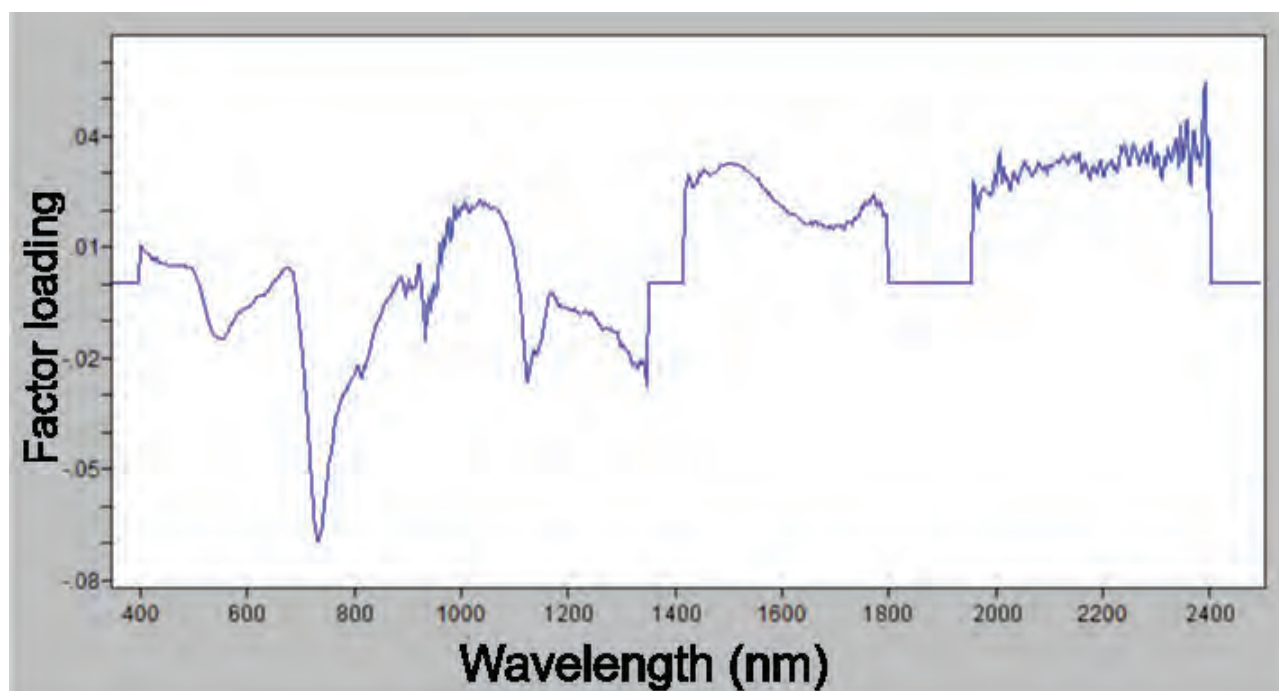


Figure 1. Spectral principal factor loadings in the spectral range related to tall fescue N concentration.

Greenseeker with rising plate

Field data from the mobile rising plate apparatus resulted in significant ($P < 0.01$) correlation coefficients (r) of NDVI with CP and FM of 0.69, and 0.85, respectively. Since fertilizer N increased FM much more than CP, canopy density and leaf area index were likely impacted more than CP. An earlier prediction equation that factored FM and NDVI resulted in a prediction coefficient of $r = 0.88$.

Conclusion

The spectral regions of the tall fescue sward that related best to forage N (thus CP) were 1421-1600 nm and 2200-2400 nm. The spectral region(s) measured by the Greenseeker appears to include wavelengths adequate for detection of CP in tall fescue. However, expression as NDVI includes confounding effects that must be accounted for in any method to estimate CP or other specific characteristics.

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