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Technical Note: Fecal NIRS equation field validation

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Abstract

Seven trials, independent of data used to develop fecal near infrared reflectance spectroscopy (NIRS) equations, were conducted to validate previously developed fecal NIRS equations for prediction of forage diet crude protein (CP) and in vivo-corrected digestible organic matter (DOM) under field conditions. For both crude protein and digestible organic matter, strong relationships existed between conventional chemistry values of diet samples collected with esophageal-fistulated steers and NIRS predictions from fecal samples collected from intact, mature, Brahman × Hereford cows at 72 hour after grazing was initiated in trial pastures.

Key Words: crude protein, digestible organic matter, near infrared reflectance spectroscopy

Recent studies have indicated potential for using fecal near infrared reflectance spectroscopy (NIRS) analysis to predict forage diet quality of free-ranging herbivores (Brooks et al. 1984, Coleman et al. 1989, Lyons and Stuth 1992). The objective of this study was to validate forage diet quality predictions of free-ranging cattle using previously developed fecal NIRS equations (Lyons and Stuth 1992). Grazed forages represented a wide range of forage quality and included both native forage species and an introduced, complementary, cool-season forage typically used in the study region.

Study Area and Methods

This study was conducted at the Native Plant and Animal Conservancy near College Station, Tex. (30° 37' N, 96° 21' W). Native vegetation at this location was characterized (Olson 1984) as having a post oak (*Quercus stellata* Wang.) overstory with a herbaceous component dominated by little bluestem (*Schizachyrium scoparium* Michx.) and brownseed paspalum (*Paspalum plicatulum* Michx.). Available vegetation was represented by a broad array of species comprised of perennial and annual C_3 and C_4 grasses and forbs as well as deciduous and evergreen woody species. All major phenological stages were represented in the study.

Seven trials were conducted in August, September, and December of 1990 and in February, March, April, and July of 1991. All trials were independent of data used to develop NIRS calibration equations (Lyons and Stuth 1992). Trials were conducted in 4 different native pastures and 1 fertilized Gulf Coast ryegrass (*Lolium perenne* L.) pasture. Average pasture size was 4 ha.

In each trial, forage diet samples were collected using 3, nonfasted, esophageal-fistulated steers. Following diet sample collection, eight non-cannulated, mature Hereford × Brahman cows grazed trial pastures for 72 hours. Fecal grab samples were taken from these cows as they entered trial pastures (0 hour) and at 12-hour intervals from 24 to 72 hours. Both esophageal-fistulated steers and intact cows had been in residence on adjacent pastures for 1 year before beginning this study.

Extrusa samples were analyzed for percent crude protein (CP) on a dry matter basis (Hach 1987) and digestibility (Tilley and Terry 1963, Van Soest and Wine 1967). Duplicate digestibility estimates were obtained for each steer sample and expressed as percent in vivo-corrected digestible organic matter (DOM) as described by Lyons and Stuth (1992). Fecal samples were stored frozen, dried in forced-air ovens at 60° C, and moisture stabilized (Lyons and Stuth 1991) before NIRS scanning. Fecal NIRS spectra were used to predict diet crude protein and digestible organic matter using fecal NIRS equations developed with cattle (Lyons and Stuth 1992).

Linear regression (SAS 1988) was used to analyze the data. The relationship between mean diet wet chemistry analyses and fecal NIRS predictions for individual cows was determined for each fecal collection period from 0–72 hours. Data was also analyzed according to the monitoring procedure suggested by Shenk et al. (1989) to determine the existence of 1) a significant bias and 2) a significant increase in unexplained error.

Results and Discussion

Conventional chemistry crude protein content for the 7 validation trials ranged from 5.4% to 27.1% (Table 1) and digestible

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Table 1. Mean extrusa sample crude protein (CP) and in vivo corrected-digestible organic matter (DOM) and mean 72-hour fecal near infrared reflectance spectroscopy (NIRS)-predicted CP and DOM by trial.

	CP				DOM			
Trial	Extrusa	SD[a]	NIRS	SD[b]	Extrusa	SD[a]	NIRS	SD[b]
Aug. 1990	8.6	1.15	9.6	0.49	57.4	2.63	59.9	0.46
Sep. 1990	7.6	0.12	8.1	0.45	58.1	1.05	59.6	0.74
Dec. 1990	5.9	1.24	5.3	0.77	57.3	1.25	54.9	0.24
Feb. 1991	5.4	1.98	6.3	0.87	50.4	4.61	53.8	0.43
Mar. 1991	27.1	0.90	27.3	1.56	74.1	0.67	77.0	4.60
Apr. 1991	14.4	1.96	12.7	0.69	63.9	1.32	60.2	0.20
Jul. 1991	11.5	2.62	9.5	0.55	57.0	1.70	59.4	0.27

[a]SD = Standard deviation for extrusa sample CP and DOM, n=3.
[b]SD = Standard deviation for fecal NIRS-predicted CP and DOM, n=3 selected at random.

organic matter ranged from 50.4% to 74.1% (Table 1). Lowest values for both CP and DOM occurred in February, 1991 with dormant, native forage. Highest values for both CP and DOM occurred in March, 1991 with fertilized ryegrass. The data range for the calibration samples upon which the fecal NIRS equations were based were from 4–17% and 54–67% for CP and DOM, respectively. The range of the current data therefore exceed that for the calibration samples, a rigorous test for the equations. Mean 72-hour fecal NIRS predictions ranged from 5.3% to 27.3% for CP and from 53.8% to 77% for DOM. In general, variation among extrusa samples for animals within individual trials tended to be greater than variation among fecal NIRS-predicted values for both CP and DOM (Table 1).

Using the monitoring procedure suggested by Shenk et al. (1989), observed bias between NIRS predictions and reference samples and unexplained error associated with predictions were calculated for both crude protein (CP) and digestable organic matter (DOM). Bias for both CP and DOM was within acceptable limits, while unexplained error for both was near the limits. Observed bias for CP was 0.24 compared to the calculated bias confidence limit of ±0.48. Unexplained error for CP was 1.22 compared to an error limit of ±1.17. Observed DOM bias was 0.01 compared to a ±0.90 confidence limit and unexplained error was 1.98 compared to a ±2.10 limit.

Coefficients of simple determination (r^2) for the regression of diet crude protein content and fecal NIRS predictions increased from a low (r^2 = 0.18) at the 0-hour sampling period to a high (r^2 = 0.98) at 72 hours (Table 2). Standard error of prediction (SEP) decreased from 1.25 at 0 hour to 0.49 at 72 hours. Intercept decreased from 8.2 at 0 hour to -0.1 at 72 hours and was not different from 0 at the 60-hour (p = 0.2481) or 72-hour (p = 0.7688) sampling periods. Slope increased from 0.19 at 0 hour to 0.98 at 72 hours and was not different from 1 at 60 (p > 0.100) or 72 hours (p > 0.250).

The r^2 for the regression of diet digestible organic matter and fecal NIRS predictions rose from a low (r^2 = 0.02) at 0 hour to a high (r^2 = 0.87) at 72 hours (Table 3). A decrease in standard error of prediction (SEP) (Table 3) occurred from 0 hour (1.60) to 48 hours (0.89). Intercept declined from 55.2 at 0 hour to 2.4 at 72 hours, and was not different from 0 at 48 (p = .1771), 60 (p = 0.6090), or 72 hours (p = 0.8157). Slope increased from 0.07 at 0 hour to 0.97 at 72 hour and was not different from 1 at the 48 (p > 0.100), 60 (p > 0.250), or 72-hour (p > 0.250) sampling periods.

Considering all parameters for crude protein (CP) (r^2 = 0.98, SEP = 0.49, intercept = –0.1, slope = 0.98) and digestible organic matter (DOM) (r^2 = 0.87, SEP = 1.12, intercept = 2.4, slope = 0.97), the 72-hour sampling period appears to provide the closest relationship between diet CP and DOM and NIRS-predicted CP and DOM. Although intercept and slope were not different from 0 and 1 at 60 hours and 48 hours for CP and DOM predictions, respectively, probabilities for these hypotheses were greatest at the 72-hour sampling period.

Trends in fecal NIRS predictions by collection period are shown for 1990 and 1991 trials in Figure 1. Because of similarity, August 1990 and July 1991 were combined into summer trials and December 1990 and February 1991 trials were combined into winter trials. Also in Figure 1, the April 1991 trial is designated as spring, the September 1990 trial is designated as fall, and the March 1991 trial is designated as ryegrass. The most dramatic change in predictions from the 0 hour to 72 hours was in the rye-

Table 2. Mean extrusa sample crude protein (CP) versus mean fecal near infrared reflectance spectroscopy (NIRS)-predicted forage CP by sampling hours across the 7 validation trials.

Hour	r^2	SEP[a]	Intercept	Probability[b]	Slope	Probability[c]
0	0.18	1.25	8.2	0.0200	0.19	<0.005
24	0.53	1.06	7.0	0.0190	0.36	<0.005
36	0.88	0.68	4.7	0.0159	0.60	<0.005
48	0.97	0.49	2.2	0.0618	0.82	<0.025
60	0.96	0.57	1.5	0.2481	0.89	>0.100
72	0.98	0.49	-0.1	0.9283	0.98	>0.250

[a]SEP = standard error of prediction.
[b]Probability that the intercept is not different from 0
[c]Probability that the slope is not different from 1.

Table 3. Mean extrusa sample in vivo-corrected digestible organic matter (DOM) versus mean fecal near infrared reflectance spectroscopy (NIRS)-predicted forage DOM by sampling hour across the 7 validation trials.

Hour	r^2	SEP[a]	Intercept	Probability[b]	Slope	Probability[c]
0	0.02	1.60	55.2	0.0118	0.07	<0.005
24	0.29	1.39	43.2	0.0172	0.29	<0.010
36	0.75	1.01	26.4	0.0320	0.58	<0.025
48	0.87	0.89	14.9	0.1171	0.77	>0.100
60	0.87	1.10	5.3	0.6090	0.92	>0.250
72	0.87	1.12	2.4	0.8157	0.97	>0.250

[a]SEP = standard error of prediction.
[b]Probability that the intercept is not different from 0.
[c]Probability that the slope is not different from 1.

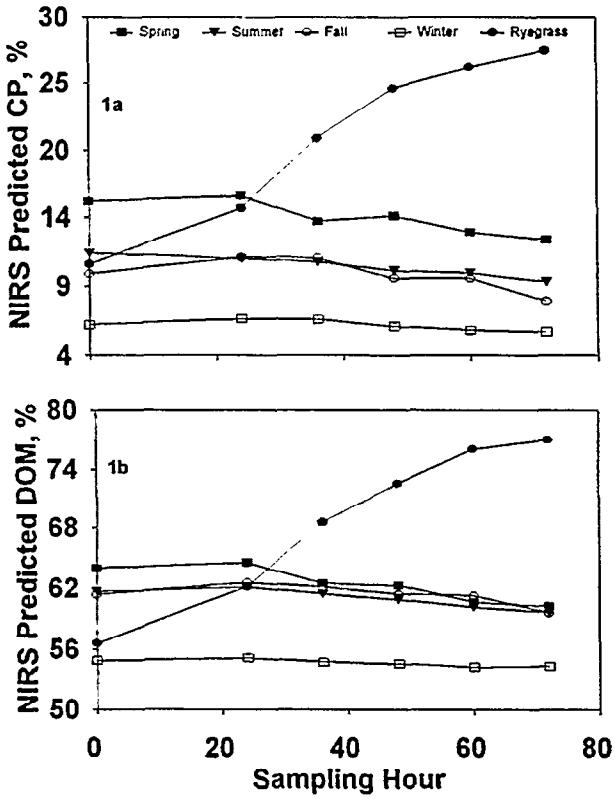

Fig. 1. NIRS-predicted crude protein (CP;1a) and in vivo corrected-digestible organic matter (DOM;1b) for Spring (April 1991), Summer (August 1990 and July 1991), Fall (September 1990), Winter (December 1990 and February 1991), and Ryegrass (March 1991) trials by sampling period. The Ryegrass trial reflects a transition from early spring native vegetation to fertilized ryegrass.

grass trial conducted in a fertilized, cool-season, monoculture. In this trial, crude protein predictions increased from about 10% at 0 hour to 27.3% at 72 hours. Likewise, digestible organic matter predictions in this trial increased from about 56% at 0 hour to 77% at 72 hours. Differences between 0 hour and 72 hours predictions for other trials were less dramatic because cows were grazing native forages before and during trials. Available forages in pastures during these trials were similar in species composition and phenological stage to pre-trial areas in which cows were grazing.

Results of this study lend further support to the feasibility of using fecal NIRS monitoring to estimate forage diet quality of free-ranging cattle. Equations tested here appear to have utility over a wide variety of herbaceous vegetation and beyond the range of data used to develop the equations. However, caution should be exercised when using these kinds of equations outside their data range and in forage types drastically different from calibration data sets. Significant deviations in forage types or quality ranges may necessitate development of specific-use equations, such as, for small grain forages with high rumen-degradable protein or for exclusive legume hay.

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