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# Fecal NIRS equations to assess diet quality of free-ranging goats

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## Abstract

Research was conducted to evaluate the predictability of the nutritional status of free-ranging goats through analysis of fecal material by near infrared reflectance spectroscopy (NIRS). Diet samples were collected from esophageally fistulated goats, whereas fecal samples were obtained from nonfistulated animals grazing the study areas. Percentage of crude protein (CP) and standard corrected in vitro digestible organic matter (DOM) were determined for diet samples. The resulting diet reference data and the fecal spectra were used to develop predictive equations. Standard errors of calibration (SEC) for CP and DOM were 1.12 and 2.02, being within acceptable limits for NIRS. Coefficients of determination ( $R^2$ ), were 0.94 and 0.93 for CP and DOM, respectively. Validation trials, performed in post oak woodlands and subtropical thornshrub regions of Texas, indicated that the selected CP and DOM equations can be useful in predicting the nutritional status of goats under different rangeland conditions.

*Keywords:* Goat; Nutrition; Fecal equation; NIRS equation

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## 1. Introduction

NIRS technology has been successfully used to predict forage quality for livestock, via esophageal extrusa (Holechek et al., 1982b), clipped forage (Barton and Burdick, 1983; Park et al., 1983; Marten et al., 1984), and livestock performance (Eckman et al., 1983). Recent studies by Coleman et al. (1989), Stuth et al. (1991), Lyons and Stuth (1992) have shown the potential use of NIRS to predict diet quality of free-ranging cattle through fecal analysis. Fecal NIRS profiling has been linked with nutritional management models to provide a comprehensive decision support system for cattle producers (Ranching Systems Group, 1993). Application of fecal prediction equations developed by Lyons and Stuth (1992) for cattle were un-

successful for goats, indicating that goat feces are biochemically different from cattle feces.

Fecal material is composed of undigested diet plus microbial matter, metabolic excretions (Van Soest, 1982), and includes plant cuticular waxes, aromatic compounds, indigestible cellulose, microbial bodies, and endogenous secretions tissue. Since different species of livestock have different diet selectivity and digestive physiology (Hanley, 1982; Van Soest, 1982; Huston et al., 1986), fecal NIRS equations for one species may not be applicable for another, essentially due to spectral diversity. Consequently, the objective of this research was to develop and select fecal NIRS equations to predict diet quality (crude protein and digestibility) of goats under free-ranging conditions.

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## 2. Material and methods

### Field area

To provide data for calibration of NIRS and development of a master equation for diet quality prediction for goats, a study was conducted on the Texas A&M Native Plant and Animal Conservancy, approx. 8 km west of College Station (30.37 N, 96.21 W, 100 m above sea level). The area is representative of the Claypan Savannah region of Texas, being dominated by an overstory of post oak (*Quercus stellata*). Yaupon (*Ilex vomitoria*) is a dominant shrub, and the herbaceous vegetation includes several grass and forbs species (Gould, 1975). The climate is subtropical with hot summers and mild to cold winters. Total annual precipitation averages 940 mm and varies from 780 to 1100 mm, and mean temperature ranges from 10°C in January to 30°C in July (US Department of Commerce, 1990).

### Field methods

Extrusa and fecal samples were collected from adult (24 to 36 mo) Spanish goats weighing 28 to 35 kg. Extrusa samples were collected from esophageally fistulated nannies, while fecal samples were collected from non-fistulated mutton goats serving as grazers on the same plots.

Samples were collected from five small paddocks, each simulating a particular kind of predominant vegetation type (native grasses, evergreen shrubs, deciduous shrubs, cool-season grass, and a grassland savanna). The calibration trials were conducted in September and November of 1990, and February, April, June, and August of 1991, each lasting 5 days.

### Laboratory procedures

All extrusa samples were dried in a forced-air oven at 60°C for 48 h. Dried samples were ground in a Udy mill to pass a 1-mm screen to reduce particle size. Esophageal samples were analyzed for crude protein (CP) by micro-Kjeldahl procedure using the Hach system (Hach Co., 1990). Digestible organic matter (DOM) was determined by in vitro procedures in samples after 48-h fermentation (Tilley and Terry, 1961) followed by the neutral detergent fiber (NDF) procedure (Van Soest and Wine, 1967). Forty-eight h in vitro values were corrected to known in vivo values (Hunt et al., 1990) by regression equations similar to

the method reported by Holechek et al. (1986), as adapted and described by Lyons (1990). The time of correction for in vivo values was assumed to be 56 h, representing a maximum value of resident time in goats' digestive tract (Huston, 1991). The CP and DOM values of the samples collected from each treatment were averaged for each day across animals for use as reference values in NIRS equation development.

Frozen fecal samples were dried at 60°C in a forced air convection oven for 48 h and then ground in a Udy mill to pass a 1-mm screen. Before scanning, samples were again placed in the oven at 60°C for 12 h to stabilize moisture (Lyons and Stuth, 1992). When removed from the oven, the samples were placed in a desiccator for 1 h to cool to ambient temperature (Abrams, 1985). After being removed from the desiccator, samples were packed in sample cups with quartz lenses. After being packed the samples were immediately scanned with a Pacific Scientific NIR Scanner 4250. The spectra generated were stored in a micro-computer interfaced with the NIR scanner for use in generation of prediction equations.

### Equation development

NIRS involves a calibration procedure requiring both the near infrared reflectance spectra of the reference material and reference data related to the variables to be predicted from spectral information (Hruschka, 1987). To match the fecal spectra of each sample with the reference data, CP and DOM dietary data for d 2 and d 3 of a trial were averaged, and the result was used as reference data for d 3 fecal samples. Mean reference data for d 3 were added to the value of d 4 and the average was used as reference data for d 4. The same procedure was used to calculate reference data for d 5. Due to a pre-condition stage, no fecal samples were collected in d 1 and 2.

Dietary CP and DOM were used as dependent variables, while stored NIRS spectra from fecal samples were used as independent variable reference data for calibration equation development. Equations were developed by modified stepwise regression, which selects the best combination of fitting wavelengths (Westerhaus, 1985a). The first term of the regression equation is simply the best fitting wavelength. The second term is one member of the best fitting pair whose other member is fixed as the first term. The second term is then fixed, and an attempt is made to find a term that

fits better than the original first term. Each member of the pair is rejected in turn until no further improvements are found. These two terms are fixed as an attempt is made to find a good third term. Then each term is rejected one at a time as an attempt is made to find a better set of three terms. The same procedure is then extended to find sets of four or more terms (Westerhaus, 1985a).

The stable equations were determined after the elimination of possible outliers and data being subjected to various statistical treatments (Williams, 1987). For each statistical treatment the best equations were identified through the consideration of several factors which include the standard error of calibration (SEC), laboratory standard error (SEL) (Westerhaus, 1985a), coefficient of determination ( $R^2$ ) (Neter et al., 1989),  $F$  value (Westerhaus, 1985b), wavelength coefficient magnitude (Williams, 1987), and examination of wavelengths to determine the existence of chemical relationships with the parameters being studied (Westerhaus, 1985b). To avoid multicollinearity problems, equations that present one or more terms with  $F$  value lower than 10 must be rejected. Also, equations with coefficients exceeding 10 000 were rejected.

#### *Equation validation procedure*

Once an equation is selected, it should be validated with unknown samples which come from trials different from those used to build the model, i.e., through the collection of new data (Westerhaus, 1985b). To validate the fecal NIRS equations developed for predicting CP and DOM, fecal and dietary samples were collected from validation trials developed at College Station and the La Copita Research Area, approx. 20 km southwest of Alice, TX, and 400 km southwest of College Station.

Validation samples were collected from three trials (October 1991, December 1991/January 1992, and April 1992) at College Station, in the same paddocks where the samples to build the equations were collected. At the La Copita Research Area, validation trials were conducted in August, October, and December of 1990, and monthly from February to August of 1991. Samples were collected from six, 1.8 ha paddocks, reflecting three levels of available browse (low, moderate, and high), replicated twice.

Ecological and climatological conditions in South Texas are distinctly different from those found in College Station, where the equations were developed. The

area is characterized by dense thornshrub dominated by mesquite (*Prosopis juliflora*) and a complex of over 20 shrubs, 27 grasses, and 52 forb species (Hanson, 1987). The climate is subtropical with hot summers and mild winters. Mean temperatures range from 13°C in January to 29°C in August. Total annual precipitation averages 724 mm. Rainfall pattern is bimodal, with peaks in late spring (May–June) and early fall (September) (National Climatic Center, 1983).

### **3. Results and discussion**

#### *3.1. Crude protein equation*

The CP equation was developed from a subset of samples with values ranging from 4.3 to 25.1% CP. After elimination of suspect samples due to wet chemistry errors and contamination of fecal extrusa as well as sample clusters with similar values (Williams and Norris, 1987), the selected equation was based on a 163-sample calibration set. The coefficient of determination ( $R^2$ ) was 0.94 (Table 1), which was similar to values reported by other authors (Holechek et al., 1982a; Brooks et al., 1984; Lyons and Stuth, 1992), who studied cattle data. Standard error of calibration (SEC = 1.12) (Table 1) was close to the laboratory standard error (SEL = 0.91), being within the acceptable limits for NIRS calibration procedures (Hruschka, 1987). These findings indicate that procedures used in sample preparation introduced little error. In general the SEC for CP in this study was close to those reported by other authors (Holechek et al., 1982b; Brooks et al., 1984; Lyons and Stuth, 1992).

Standard error of validation corrected for bias, SEV(C), was 1.28 (Table 1), indicating a high degree of precision in estimates. The SEV(C) is obtained by using an equation developed from odd-numbered samples predicting even-numbered samples (Norris et al., 1976). The relationship between reference CP values (lab values) and NIRS predicted values is shown in Fig. 1.

#### *3.2. Digestibility equation*

The DOM equation was developed with samples ranging from 40.9 to 71.8 percent DOM. The selected equation consisted of a 86-sample calibration set. The

Table 1

Crude protein (CP) and in vivo corrected digestible organic matter (DOM) equations for free-ranging goats from the College Station calibration set

Equation	Calibration					Validation			
	<i>n</i>	wavelength	<i>F</i>	SEC	<i>R</i> <sup>2</sup>	SEV(C)	<i>r</i> <sup>2</sup>	bias	slope
CP	163	2027	170	1.12	0.94	1.28	0.94	0.16	1.18
		2174	332						
		2241	229						
		2260	124						
		2305	360						
DOM	86	2018	182	2.02	0.93	2.12	0.92	0.18	0.91
		2057	111						
		2143	89						
		2301	169						

Math treatment in both equations is 2,10,10,1 (2nd derivative of log (1/*R*) spectra).

SEC, standard error of calibration; SEV(C), standard error of validation corrected for bias; *R*<sup>2</sup>, coefficient of determination; *r*<sup>2</sup>, coefficient of simple correlation; DOM, kg digestible OM/kg DM.

lower sample size of the DOM calibration set was due to a higher degree of value clustering. *R*<sup>2</sup> was 0.93 (Table 1), which was higher than values found for cattle equations by other authors. Holechek et al. (1982b) and Brooks et al. (1984) found, respectively, *R*<sup>2</sup> values of 0.84 and 0.88 for in vitro dry matter digestibility (DMD). Lyons and Stuth (1982) reported a *R*<sup>2</sup> value of 0.80 for DOM.

Standard error of calibration (SEC = 2.02) was similar to values reported for digestibility estimates in other studies (Holechek et al., 1982b; Brooks et al., 1984; Lyons and Stuth, 1992). The SEL for DOM in this study was 1.98, indicating that the SEC is within the limits for NIRS calibration procedures. The higher relative CP standard error of calibration compared with that for DOM equation may be related to variations in the supply of nitrogen from rumen recycling and endogenous nitrogen (Lyons and Stuth, 1992).

The standard error of validation for the DOM equation was 2.12, which indicates a high degree of precision in estimates. The relationship between reference DOM values and NIRS predicted values is illustrated in Fig. 1.

### 3.3. Wavelengths examination

The final step for selection of NIRS equations involves the examination of wavelengths to determine whether meaningful chemical relationships exist for the

variables being measured (Hruschka, 1987). Although wavelengths of multiterm equations are so independent that interpretation of individual wavelengths is often difficult, it has been recommended that examination of only the first two wavelengths in terms of *F*-statistics rank should be conducted (Norris et al., 1976; Windham et al., 1988). However, because a tilting filter instrument was used in this study, only the primary wavelength was used for analysis, as suggested by Lyons and Stuth (1992).

NIR instruments determine CP, DOM, and other components by measuring log (1/reflectance (*R*)), which is related to absorption (Hruschka, 1987). A higher log (1/*R*) value means that more radiation has been absorbed (less reflected by the sample at that wavelength). To accentuate spectral characteristics, Hruschka (1987) suggested the conversion to second derivative of log (1/*R*) spectra of fecal samples representative of forage quality at extremes of a data set.

Spectra of fecal samples representing diet quality extremes for CP and DOM in this study are illustrated in Fig. 2. For the CP equation, it shows greater absorbance for the high-quality sample at the primary wavelength (2305 nm). Lyons and Stuth (1992) suggested that the greater absorbance associated with feces from high-quality forages may indicate detection of microbial response to diet quality through absorbance associated with chemical bonds in undigested rumen microbial cell wall, whole microbial cells produced in

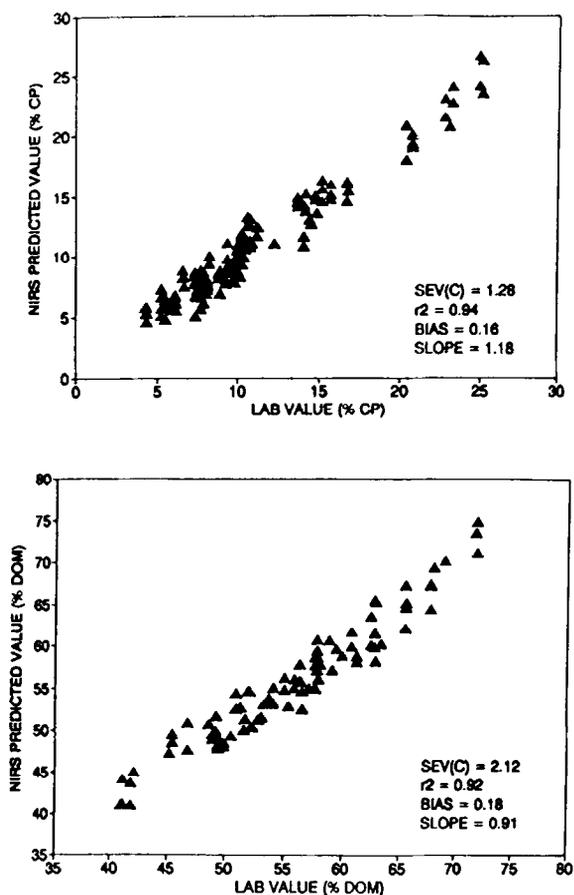


Fig. 1. Reference crude protein (CP) vs. NIRS predicted CP and reference in vivo corrected digestible organic matter (DOM) vs. NIRS predicted digestible organic matter for College Station calibration set.

the lower gastrointestinal tract, and aromatic and other by-products of microbial degradation. Norris et al. (1976) and Redshaw et al. (1986) associated wavelengths around 2305 with nm NDF of forage samples. Huston and Pinchak (1991) described CP as one of the components of NDF in plant cell wall, normally the less digestible portion of the plant cell, usually related to fecal components. Flinn et al. (1992) suggested that cuticular waxes from plant origin, such as alkanes, which are highly indigestible, absorb strongly in this region.

For the spectral region around the primary wavelength (2018 nm) in the DOM equation, absorbance was greater in the low-quality sample (Fig. 2). This wavelength falls in the range of wavelengths related

with  $-OH$  (hydroxyl) chemical bonds, which are reported to be in all starch- and cellulose-containing substances (Murray and Williams, 1987). This sample was collected in February 1991 in a paddock dominated by mature grass. It has been widely reported that digestibility of forages declines with maturity, which is associated with increased fiber or cell wall contents (Holmes, 1980; Holechek et al., 1989; Huston and Pinchak, 1991). On the other hand, the secondary wavelength (2301 nm) for DOM equation was almost as important as the primary wavelength (Table 1). In this wavelength the higher absorption was observed in the higher quality diet, which is similar to results reported by Lyons and Stuth (1992).

### 3.4. NIRS equations validation

For both CP and DOM, the standard error of predictions (SEP) found for College Station and La Copita validation data sets were similar. All SEPs were within acceptable limits (Westerhaus, 1985b) and, more important, they showed considerable improvements compared with the original equations (Figs. 3 and 4). This indicates that standard errors of prediction are not seriously biased, giving an appropriate indication of the predictive ability of the models. Even though the coefficients of determination ( $R^2$ ) were lower than those observed in the original equations, they indicate

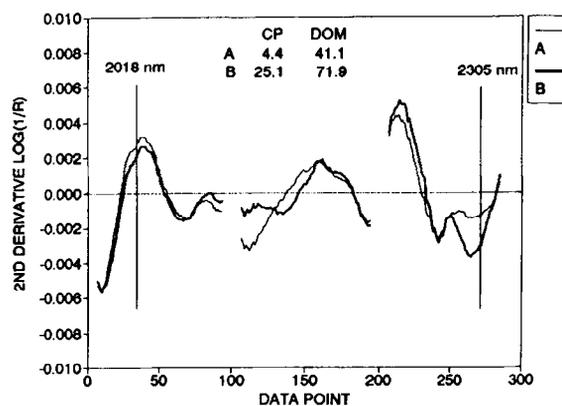


Fig. 2. Comparison of second derivative  $\log(1/R)$  fecal spectra associated with low (A) and high (B) quality forages illustrating greater absorbance at most significant estimated wavelengths in the DOM equation (2018 nm) and in the CP equation (2305 nm) for sample B. Valleys (minima) in second derivative are analogous to peaks (maxima in  $\log(1/R)$  spectra).

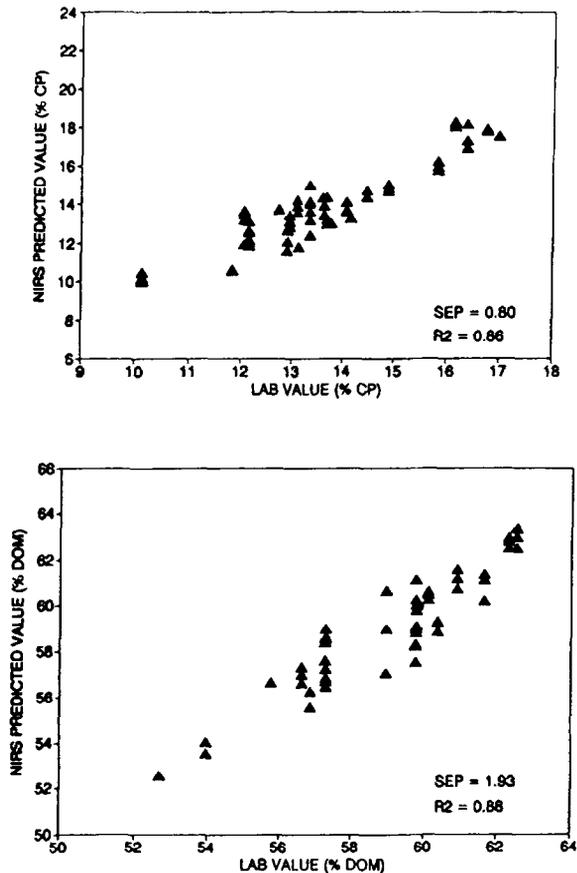


Fig. 3. Reference crude protein (CP) vs. NIRS predicted CP and reference in vivo corrected digestible organic matter (DOM) vs. NIRS predicted DOM for College Station validation set.

that a high percent of variability in the predicted values was due to variations in the independent variables (extrusa in vitro DOM) of the selected equations.

These validation analyses have shown that the selected CP and DOM fecal NIRS equations can be an important tool in predicting nutritional status of goats under different rangeland conditions. Even though climatic and botanical characteristics in the Post Oak Savannah and in South Texas are distinctly different, validation analyses for equations in both areas revealed similar accuracy. A broader application of the equations, however, would be dependent upon validations for other range conditions. Further studies would be useful for broadening the application of the equations to other regions.

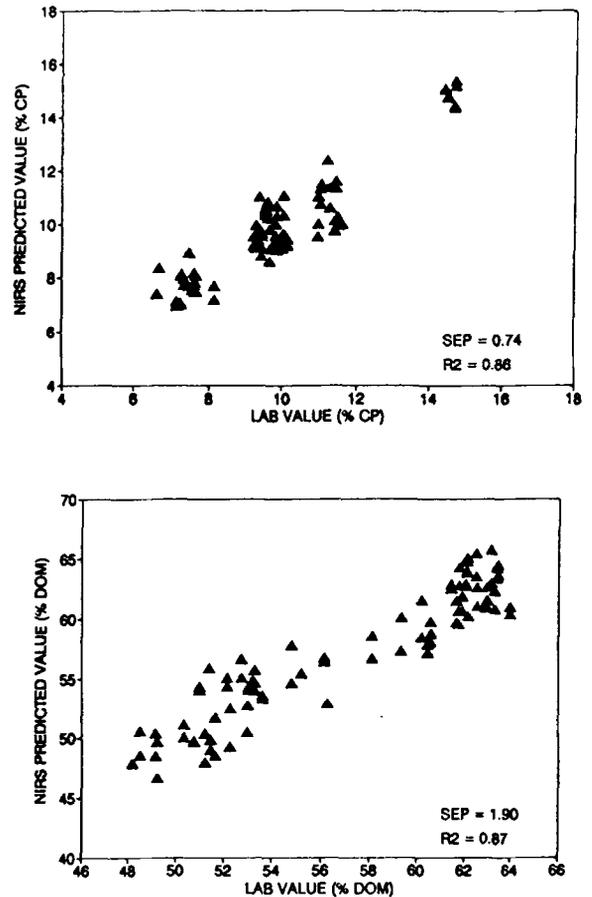


Fig. 4. Reference crude protein (CP) vs. NIRS predicted CP and reference in vivo corrected digestible organic matter (DOM) vs. NIRS predicted DOM for La Copita validation set.

#### 4. Conclusions

Previous studies have shown that NIRS has the potential to serve as a tool to reduce time, labor, and cost inputs associated with nutritive evaluation of range animal samples, helping to establish programs of animal supplementation and improving production and reproduction management systems. The present study indicates that NIRS is a viable technology for predicting diet quality of free-ranging goats. Generalized fecal NIRS calibration equations can accurately predict dietary CP and DOM of goats grazing a wide variation in botanical composition. The precision of both CP and DOM equations matched that of the conventional lab-

oratory methods. Validation of equations also showed the degree of precision in predicting CP and DOM. Similar SEP and  $R^2$  for predicted values for two different regions seem to show the accuracy of the models for a broad application.

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