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# **Appendix B**

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Analysis of the Use of the Texas A&M Grazinglands Animal Nutrition Laboratory Fecal Near Infrared Reflectance Spectroscopy Prediction Equation with California Annual Rangeland Forages Analysis of the Use of the Texas A&M Grazinglands Animal Nutrition Laboratory Fecal Near Infrared Reflectance Spectroscopy Prediction Equation with California Annual Rangeland Forages

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ANGELA DAWN JINKS B.S. (Purdue University) 2001

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## **Chapter I**

1

#### Use of Fecal Near-Infrared Reflectance Spectroscopy with California's Annual Rangeland System

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#### Introduction

California rangelands cover approximately 62 million acres. About 20 million of these acres are held privately, with approximately 40 million total acres of public and private grazing land (FRRAP, 1988). Although a significant portion of the state is rangeland, relatively little is known about California's rangeland system. The state has a unique rangeland, composed mostly of non-native annual species which germinate with fall rains (Hart et al., 1932d; Pitt and Heady, 1978). The predominance of annual species, along with a Mediterranean climate, makes research and knowledge from other systems difficult to apply in California.

In contrast to most of the United States, California receives most of its rainfall during the late fall and winter. Fall rains and warm temperatures initiate germination and development of annual grasses, which slows with cooling temperatures. Warmer spring temperatures accelerate growth, but when the rainy season ends in the spring, the annual grasses characterizing much of the state's rangelands mature and senesce (George et al., 1985b; Young et al., 1973). The summer dry season, which varies in length and timing, is of special interest to ruminant livestock producers and scientists, because inherent in the process of senescence is an increase in the fiber content of the forage leading to an increase in its bulk, which contributes to rumen fill and a decrease in energy intake. There is a further decrease in the nutritional value of these forages due to shatter and bleaching. Of particular concern for many cattle producers is the loss of crude protein (CP), which is necessary for growth, lactation and gestation in grazing cattle (George et al., 2001b). Supplementation with higher nutrient density forages, grains and/or minerals is often needed to maintain acceptable animal production levels.

To reduce costs, many beef cattle producers try to match the beef production cycle with the available forage resources. The pattern of annual forage growth was described by Bentley and Talbot in 1951, and divided into three seasons of availability for cattle: (a) the inadequate green season, (b) the adequate green season, and (c) the inadequate dry season. The inadequate green season begins with germination and extends to the beginning of the warm spring weather. Although nutritional quality of this new forage is high, both height of forage and the absolute amount available may limit voluntary intake by grazing cattle. In addition, remaining dry forage from the previous year is of very low nutritive value due to shatter and leaching of nutrients by rainfall. Warmer temperatures spur rapid spring growth, which characterizes the adequate green season. During this period, there is sufficient quantity and quality of forage to meet the nutritional demands of grazing ruminants. The inadequate dry season begins when soil moisture declines to a point where annual forages mature and senesce. With this maturation process comes a decline in nutritional quality and palatability that prevents cattle from consuming enough forage to meet their nutritional needs. Quality continues to decline throughout the dry season due to bleaching, shatter and decomposition, although the extent of the quality decline varies among forage species and location.

Most California cow-calf producers choose to calve their herds in the fall, which balances the greatest nutritional demands of the cow and calf with the inadequate and adequate green season. Feed supplements are provided as necessary, especially through the breeding season. Calves are weaned as forage quality declines with the onset of the inadequate dry season, and cows are maintained throughout the summer on standing dry forage and, in some locations, feed supplements. For a variety of reasons, such as

availability of later maturing forages or irrigated pasture, seasonal disease prevalence, such as foothill abortion, or more severe winters, some producers choose to calve their herds in the spring (Oltjen, 1982).

The varied topography, climates, soils and degree of human influence throughout the state has resulted in a collection of highly diverse rangelands. This diversity is increased by natural yearly variation in rainfall, temperatures, soil moisture, and other environmental factors (Hart et al., 1932c; Pitt and Heady, 1978). In addition, California's moderate climate allows invading species from around the world relatively easy colonization. Plant species have different nutritional values for cattle, and are available for grazing in different amounts throughout the year. Forage nutritional value also varies within forage species according to stage of maturity.

For cattle producers, nutritional management of their cattle is further complicated by selective grazing. Different classes of cattle graze differently and, even within class, individuals graze differently. On these highly diverse rangelands, predicting nutrient intake by any group of cattle, and their resulting nutritional status, has been challenging and ultimately unsatisfactory.

A number of techniques have been developed in the range and animal sciences to help alleviate this challenge. Cattle producers and their advisors routinely clip pasture forage samples and have them analyzed for their nutrient profile. The ability of a collected forage sample to reflect the available feed depends on the knowledge of the person taking the sample as well as their sampling technique and, at best, measures nutrients available rather than nutrients consumed. Still, results can help cattle producers determine the most appropriate supplementation strategies. Alternatively, producers can

use cattle performance to determine when to supplement, but this is costly in terms of production due to often late intervention with supplements. Cattle will be more susceptible to disease, wean lighter calves, and may not rebreed if not supplemented correctly. In addition, replacing lost body condition often requires more supplement than if the cattle were maintained at the appropriate nutrient level throughout the feeding period (George et al., 2001b).

#### **Near-Infrared Reflectance Spectroscopy**

#### Background

To enhance the management of rangeland beef cattle, researchers have used fecal pats as indicators of what was consumed by grazing cattle. In 1988, researchers in the Ranching Systems Group in the Department of Rangeland Ecology and Management at Texas A&M University in College Station, Texas, began investigating the use of fecal near-infrared reflectance spectroscopy (NIRS) to determine percent CP and digestible organic matter (DOM) of rangeland forages. Since 1994, the Grazingland Animal Nutrition Laboratory, or GAN Lab, has offered commercial NIRS services of fecal pats (Eilers, 1999) to predict CP, DOM and phosphorus levels of the forages consumed by grazing ruminants.

NIRS has been used for a variety of applications in science. For example, in animal agriculture, fecal NIRS has been used to determine the percent of leafy spurge in diets selected by sheep and goats (Walker et al., 1998), measure the amount of lignin to aid in determining diet digestibility (Purnomoadi et al., 1996), and predict diet nutritional quality (Leite and Stuth, 1995a);(Lyons and Stuth, 1991; Lyons and Stuth, 1992a).

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NIRS offers many benefits to scientists and producers. Unlike traditional laboratory analyses, NIRS is a non-destructive method that produces no chemical waste while yielding quantitative results. With a small sample size, any number of constituents for which prediction equations exist can be rapidly analyzed. A particular strength of NIRS over other methods of diet analysis, especially for heterogenous range diets, is that fecal NIRS measures constituents in the feces, which is a direct result of the actual diet consumed by the grazing animal. Unlike most other research methods, NIRS also allows investigation of a particular variable without artificially manipulating or separating it from the system (Coates, 2000; Foley et al., 1998). It is also a rapid method that returns information to producers soon enough to make timely nutritional management decisions. The GAN Lab advertises a 5 d response time from fecal sample collection by the producer to the time the producer receives the results. This allows for more exact nutritional management, resulting in more efficient cattle production, reduced costs of supplementation due to reduced, over, or improper supplementation, and better control over release of minerals into the environment from fecal and urine excretion.

#### Methodology/Technology

In the NIRS method, samples are irradiated with light from the near-infrared region. This specifically refers to radiation from 700 to 3000 nm, but most applications of NIRS include wavelengths from 1000 to 2600 nm (Hruschka, 1987). Near-infrared radiation is absorbed mainly by C-H, N-H and O-H bonds. As the sample is irradiated, bonds within the sample vibrate, causing the bond to stretch and bend. This stretching and bending creates a wave motion within the bond at a frequency that is specific for that functional group at that wavelength. If the frequency of the light matches the vibrational

wave of the bond, light is absorbed. Light that does not match the vibrational wave is reflected and measured by the spectrometer (Foley et al., 1998).

The intensity of the reflected light is portrayed on the vertical axis as the log of the inverse of reflectance, or log (1/R). As more radiation is absorbed at a particular wavelength, a higher log (1/R) value results (Hruschka, 1987).

In contrast to other forms of spectroscopy, the spectrum peaks of NIRS are blunt because they consist of overtones and combinations from primary absorption, mostly in the mid-near-infrared region, and because some light is scattered. There are few, if any, regions in the near-infrared region where absorbance is due to only one type of functional group (Foley et al., 1998). The optical spectra of each sample are converted to a numerical format using regression equations. To account for scatter and overlapping of constituents, multivariate equations are produced from a group of samples with known chemical composition. These samples, known as the calibration set, are paired with their near-infrared spectra to "train" the computer program to recognize similar unknown samples. The accuracy of future predictions depends heavily on the accuracy and precision with which the values for the calibration set were determined. To achieve a robust model, the calibration set should include samples with a range of spectral variation representative of the range of spectral variation seen in the population, so that the prediction equation can accurately predict future unknown samples (Marten and Naes, 1987).

Baseline variations and overlapping absorption bands often make interpretation of spectral outputs difficult. To solve these problems, various orders of the derivatives of spectra may be collected, although the second derivative is generally the highest

derivative used in practice. Higher derivatives, while resolving overlapping absorption bands, tend to be more sensitive to unexplained variation and therefore generate more artifacts than do lower order derivatives (Hruschka, 1987).

Particle size, moisture and compression of the sample within the sample cup play important roles in obtaining clear and repeatable spectra. Water is a strong absorber of light, so moisture in a sample will produce obvious absorption bands. Variations in particle size are reflected by changes in the amount of radiation scattered by the sample. Larger particles absorb more radiation than smaller particles because the direction of the radiation is not changed as often, and so they exhibit a larger log (1/R) value. Sample compression can also effect spectra, and increase variation among samples, due to differences in compression and spreading within the sample cup (Foley et al., 1998; Hruschka, 1987).

NIRS is a secondary predictor of diet quality and, as such, includes errors made during laboratory analysis of the calibration set as well as NIR instrument errors of both the calibration set and unknown samples (Coates, 2000). Still, a good NIRS prediction equation should have a prediction error similar to the error of the laboratory reference set (Foley et al., 1998).

Once calibration equations have been established and validated, NIRS can be used as a management tool. For example, the rapidity of fecal pat analysis with NIRS allows cattle producers to know the nutrient composition of their cattle's diet within 5 d of fecal sampling. This information can be used to make a variety of management decisions including whether supplementation is necessary and, if so, what and how to supplement, whether to wean early, move between pastures, or market cows. This rapid sample

analysis yields information faster than more traditional indicators of deteriorating diet quality, such as lost body condition, and so allows more efficient and timely nutritional cattle management.

#### **Equation Development**

To develop prediction equations, diets of known nutrient composition must be paired with their corresponding fecal samples. Accurately determining the nutrient composition of the diet that cattle consume on rangeland is very difficult and therefore, it is difficult to know the composition of that diet without using complex, expensive and invasive procedures that are unsuitable for commercial production situations.

Nevertheless, various methods have been used to determine diet composition or its attributes. On homogenous or monoculture pastures, clipping and analyzing forage samples may be adequate. However, western rangelands present problems due to the diversity of species present, as well as the diversity of their phenological stages and the nutritive quality within each species and stage. In much of the published literature, diet samples for equation compilation and/or validation were collected from esophageally fistulated cattle grazing with or before the resident cattle from which fecal samples were collected (Coates, 1800; Leite and Stuth, 1995b; Lyons et al., 1995; Lyons and Stuth, 1992b). However, others have found that samples collected via esophageal fistula do not represent the total mean diet consumed by the fistulated cattle (Coates et al., 1987; Jones and Lascano, 1992).

According to Arnold (1962), sheep have between 8 and 10 grazing events/d. Coates et. al. (1987) attributed differences between extrusa samples and the true diet, as determined with natural carbon isotopes, to the inability of one or two extrusa samples to represent the 8 to 10 grazing events of the day. McManus (1980) found that diet selection changes with increasing satiation, as well as changes in pasture and weather, further supporting the idea that one or two extrusa samples cannot represent the mean daily diet of the esophageally fistulated cattle. The same studies (Coates et al., 1987; Jones and Lascano, 1992) also found that when roving fistulated animals are used, they did not consume the same diet as the resident animals from which fecal samples were collected. As such, these diets would also differ in constituents such as CP, fiber, and minerals.

Two predictive equations were developed by the GAN Lab at Texas A&M using data sets from rangelands in Texas, the Midwest and the southern portion of the prairie provinces of Western Canada. Diet-fecal pairs for many of the included data sets were constructed using fistulated animals. These equations are commonly called the "cool season equation" and "warm season equation," where the cool season equation is used to estimate nutrient composition of pastures including introduced small grain forages such as rye, wheat and brome grasses. This equation is best suited for intensively managed pastures, particularly monocultures. The incorrectly named "warm season" equation is used for ranges where native C3 and C4 forages occur, such as more extensive rangeland systems. Some cool season grasses are also included in this group (D. Tolleson, GAN Lab, Texas A&M University, TX, personal communication).

The forage species growing in the predominant cattle producing areas of California are generally annual non-native grasses and forbs that do not neatly fit into either the cool or warm season designation. Even in areas of the state that boast a more perennial system, many of the grass species present differ from those found in areas

represented by the existing NIRS equations. Cattle producers in California are currently submitting samples to the GAN Lab, even though the GAN Lab equations were not developed using forages from this production system and have never been validated for use on California's annual rangelands. California cattle producers using the GAN system have reported mixed results in terms of perceived predictive accuracy based on cattle performance.

#### Conclusion

To be useful, and to maintain accuracy and precision, NIRS equations must be derived using samples from the same population that the equation will later be used to predict. The heterogenous nature of California's rangelands, coupled with the fact that California has a unique annual rangeland consisting of many species not found on rangelands in the rest of the United States, suggest that evaluation of the existing Texas A&M GAN Lab NIRS equations, and perhaps formulation of new predictive equations, was desirable.

### **Chapter II**

#### Validation and Improvement of the Texas A&M Grazinglands Animal Nutrition Laboratory Near-Infrared Reflectance Spectroscopy Prediction Equation

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ABSTRACT: Near-infrared spectroscopy (NIRS) of fecal samples has been used to predict the CP and DOM of the forages consumed by grazing animals. However, for NIRS predictions to be accurate, the prediction equation must be based on samples from the same population as the samples to be predicted. Beef cattle in California graze a unique rangeland composed mostly of non-native annual species which germinate with fall rains. The predominance of annual species, along with a long dry summer during which annual grasses mature, die and decrease in quality, makes information from other systems difficult to apply to California rangelands. The Grazinglands Animal Nutrition Laboratory (GAN Lab) has a NIRS program based on forages in Texas, the Mid-west and the lower part of the prairie provinces of Western Canada. California's annual grasses do not fit into this system, but producers in California have been using the Texas A&M GAN Lab NIRS predictions despite the fact that those equations have never been evaluated under California conditions. Beef cattle digestibility trials on two California rangeland summer forages were conducted to produce forage-fecal pairs to test the existing NIRS equations and to develop new equations if necessary. Predictive capability was different between the sites. The difference between the laboratory CP value and the value predicted by the original NIRS equation varied from 2.02% units to 4.25% units at the two sites (P < .01). DOM was overpredicted by 4.25 and 5.14% units (P < .01) at the two sites with the original equation. The difference between the laboratory CP and DOM predictions by the new equation did not differ (P=0.95 and P=0.31 respectively). This study shows that the addition of forage-fecal paired data produced on California rangelands improves predictive ability of the GAN Lab system and reduces the overprediction of CO and DOM during the critical dry season.

#### Introduction

California has a unique rangeland composed mostly of non-native annual species which germinate with fall rains. This annual system, along with other differences, makes research and knowledge from other rangeland systems difficult to apply.

In comparison to much of the rest of the United States, California receives most of its rainfall during the late fall and winter. Fall rains and warm temperatures initiate plant germination and growth, which slows with cooling temperatures. Growth accelerates with warmer spring temperatures, but when the rainy season ends in late spring, the annual grasses characterizing much of the state's rangelands mature and senesce. The dry season is of special interest to livestock producers and scientists, because inherent in the drying process is an increase in fiber content, which contributes to rumen fill, a decrease in forage energy content, and an increase in shatter and bleaching, which causes further decreases in the nutritive value of the forage. Of particular concern for many producers is the loss of crude protein (CP), which is important for growth, lactation and gestation in cattle. To maintain acceptable levels of production, cattle producers often must supplement with forages, grains and/or minerals in various forms. Even during times when the energy available is high, energy intake is generally limited by rumen fill due to high fiber levels.

Cattle producers must determine when, how, and how much supplement to provide to meet the changing nutrient demands of their cattle. Traditional methods of determining when to supplement include tracking cow weight and/or body condition score, hand sampling of available forage, visual appraisal of rangelands and, in some cases, routine supplementation in certain seasons or at specific production stages. A tool to rapidly access nutritive quality of forages consumed would improve cattle producers' ability to determine how to best supplement their cattle.

Near-infrared reflectance spectroscopy (NIRS) has been attracting the attention of scientists in a variety of scientific fields since the late 1970's. Although the technology can be used to predict chemical composition of many different substrates, including forages, fecal spectroscopy is unique because it provides a prediction of the nutrient composition of the diet that was actually consumed by grazing cattle.

Two predictive equations were developed by the GAN Lab at Texas A&M University in College Station, TX, using data sets from rangelands in Texas, the Midwest and the southern portion of the prairie provinces of Western Canada. These equations are commonly called the "cool season equation" and the "warm season equation," where the cool season equation is used in pastures including introduced, small grain-type forages such as rye, wheat and brome grasses. This equation is best suited for intensively managed pastures such as monocultures. The incorrectly named "warm season" equation is used for ranges where native C3 and C4 forages are found, such as more extensive rangeland systems. Some cool season grasses are also included in this group (D. Tolleson, GAN Lab, Texas A&M University, TX, personal communication). The cool season equation is rarely used to predict nutrient composition of California forages, and so, was not examined in this study.

The forage species found in the cattle producing areas of California are generally annual non-native grasses and forbs that do not completely fit into either the warm or cool season designation. Even in areas of the state that boast a more perennial system, many of the grass species are different than those found in the areas represented by

existing NIRS equations. Cattle producers in California are currently using this system, despite the fact it was not developed using forages from California's unique production system and has never been validated for use in California. California cattle producers using the GAN system have reported mixed results in terms of perceived predictive accuracy based on cattle performance.

The purpose of the trial was to:

- Determine whether the Texas A&M fecal NIRS warm season equation accurately predicts forage nutrient composition of California forages during the dry season, and;
- If necessary, to develop a new NIRS regression equation to improve prediction of forage quality under California conditions.

#### **Materials and Methods**

Starting in June of 2002, rangeland forage was harvested from two California sites to provide forage for *in vivo* digestibility trials. The first site was at the Sierra Field Research and Extension Center (SFREC), located near Marysville, CA. and the second site was near Petaluma, CA.

#### Site Descriptions

**Petaluma**: The site at Petaluma was on privately owned land about 10 km west of the town of Petaluma, which is 55 km north of the Golden Gate. Forage was harvested from a relatively steep slope with a western aspect. The surrounding region is heavily influenced by coastal weather patterns, as it is located approximately 25 km from the Pacific Ocean and approximately the same distance from San Pablo Bay. Fog is common throughout the entire year. This atmospheric moisture contributes to a decline in forage quality above what occurs further inland. The area receives about 64 cm of rain/yr, primarily in the late fall to spring.

SFREC: An additional site was located at the Sierra Foothill Research and Extension Center (SFREC), 25 km east of Marysville and 95 km northeast of Sacramento, CA. Forage was harvested from mostly flat to rolling ground with an eastern exposure. The site was dominated by annual grasses and is typical of the land grazed by many cattle in California. It has a hot and dry climate, being on the eastern side of the Sacramento Valley in the Sierra foothills. Average rainfall is 71 cm/yr, with rain events generally in the late fall and spring.

The main species at SFREC in late May and early June, 2003 were Wild Oat (Avena barbada), Rose Clover (Trifolium hirtum), Medusa head (Taeniatherum caput medusae) and Soft Chess (Bromus hordeaceus). The main species at Petaluma were Annual rye (Lolium multiflorum), Ripgut brome (Bromus diandicus), Foxtail (Hordium leporanium) and Wild Oat (Avena fatua) (Table 1).

#### Harvest Protocol

To minimize shatter losses, range forage was cut using a sicklebar mower at SFREC, or a rotocombine at Petaluma, to a stubble height of approximately 10 cm. Forage was raked into rows and placed on tarpaulins for transfer to a trailer. To minimize forage species variation among harvests, the total area to be harvested was divided and a portion of each section included in each harvest. Harvest occurred at 6 wk intervals at each site, with the sites offset by 3 wks. Harvest continued at each site until one harvest following the first germinating rain, defined by 12 to 25 mm of rainfall within 1 wk (George et al., 2001; George et al., 1985; George et al., 1988; Bentley and Talbot, 1951). The first harvest at the SFREC was on June 11, 2002 and the first rain event of 40 mm occurred on November 7, with an additional 119 mm falling on November 12. The last harvest occurred at the SFREC on November 26, 2002, 2 wks following germination. Significant "green-up" was observed by the time of the last SFREC harvest. No significant rainfall occurred at the Petaluma site prior to the last Petaluma harvest. A total of 5 harvests at SFREC and 4 harvests at Petaluma were collected.

#### **Digestibility Trial Protocol**

The harvested range forage was chopped to an approximate average length of 7.6 cm to increase voluntary intake by the cattle and to minimize sorting. An average of 5 cross-bred Angus steers were fed chopped forage every 8 h to meet predicted maintenance energy requirements. Immediately prior to forage feeding, soybean meal was offered to bring total N in the total diet to 3% of DM. Water was offered *ad libitum*.

Steers were housed in individual pens at a feedlot at the University of California, Davis, which is located approximately 3 km west of Davis, CA. All animal procedures were approved by the University of California, Davis Animal Care and Use Administrative Advisory Committee.

Steers were fed for a 14 d adjustment period, followed by a 5 d total fecal collection period using fecal harnesses. Fecal samples were composited on a percentage of total fecal output basis by day, steer and period. All composite samples were preserved in triplicate and frozen. One sample was sent by 2 d mail to the Texas A&M GAN Lab for NIRS analysis, and a second sample was dried at 50°C for 72 h before being ground to pass a Wiley mill (Arthur H. Thomas, Philadelphia, PA) 1 mm screen. Dried fecal

samples were analyzed for (CP), acid detergent fiber (ADF), neutral detergent fiber (NDF), and ash by the Dairy One Forage Laboratory (Ithaca, NY). Dry matter was determined at the time of compositing by drying at 50°C for 72 h.

Forage samples were collected at chopping, and forage and soybean meal samples were collected once during the fecal collection period. Dry matter for forage and soybean meal was determined once during the fecal collection period throughout the dry season, and then once daily from day 13 to day 18, once the fall rains began, by drying at 50°C for 72 h. Significant feed refusals, if they occurred, were collected and weighed daily from day 13 to day 18, and were composited in the same manner as fecal samples. Forage, soybean meal and refusal samples were dried, ground to pass a 1mm screen and analyzed for CP, ADF, NDF and ash, as described previously. All nutrient analyses were conducted by the Dairy One Forage Laboratory (Ithaca, NY).

Forage CP was determined by standard laboratory nutrient analysis. The following equation was used to calculate digestible organic matter (DOM) of the forage. Forage DOM: (DM forage intake \* forage OM- (fecal DM output\*fecal OM – fecal OM SBM))/(DM forage intake)

where:

Fecal OM SBM = DM SBM intake \* (1-ash)\*0.85 and soybean meal is assumed to be 85% digestible (NRC, 1996)

#### Statistical Analysis

NIRS warm season equation prediction errors of day composite fecal samples were analyzed using the Proc GLM in SAS (SAS Inst. Inc.; Cary, NC) in a model that

included the effect of equation (original warm season vs. improved warm season), time (date of harvest), location (SFREC vs. Petaluma) and all interactions.

#### **Equation** Compilation

Equations for forage CP and DOM were constructed using forage-fecal pairs whose predicted constituents fell within 1.5 standard deviations of the expected value based on similar spectra from the same population of samples. An equation was developed using forage-fecal pair data from the day composites obtained from the digestibility study. Day composites were used because they match what occurs when a cattle producer takes a sample on one day from several fecal pats.

#### **Results and Discussion**

Laboratory CP values for each period at the sites show a small increase, while, DOM showed a general trend towards decreasing, as the summer progressed (Table 2.2). Overall, CP was 1% unit higher for the dry season at Petaluma than at SFREC. DOM was approximately 1% unit higher for the average of the dry season at SFREC than at Petaluma. The original and new NIRS equation prediction (Table 2.3) and prediction errors (Table 2.4) are shown for each harvest at both sites.

#### **Crude Protein**

The original equation overpredicted CP by approximately 2 and 4% units (Table 2.4) at the Petaluma and SFREC respectively (P<0.01). Addition of dry season forage-fecal pairs to the existing GAN Lab equation improved predictive capability for CP (P<.05) (Figure 2.1 and 2.2). All single factors were significant, and location by equation

and location by time interactions occurred (P<0.01). Because there was a location by equation interaction, data were analyzed by location.

Data from the Petaluma site show that both time and equation used are significant (P<0.01). On average, differences between least square means (prediction – laboratory value) (Table 2.4) demonstrates that predictive capability for CP was improved (P<0.01) by more than 2% units with the improved equation (Figure 2.5, 2.7).

At the SFREC site, equation, time and the equation by time interaction were important sources of variation (P<0.01) (Figure 2.6, 2.8). Comparison of sources of variation(F-test) showed that equation was the dominant source of variation (P<0.01) Differences between least square means shows the improved equation was more than 4% units better at predicting than the original equation (Table 2.4).

Least square means of predictions of CP at the two sites by the new equation did not differ from zero. However, the original equation more accurately predicted Petaluma samples than SFREC samples (P<0.01). The 2% to 4% unit improvement in prediction has important implications for cattle producers making nutrient management decisions for their cattle.

#### **Digestible Organic Matter**

Predictions of DOM were more accurate under the new equation than predictions with the original equation (P<0.01) (Figure 2.3, 2.4). Accuracy of predictions of DOM for Petaluma samples were 4.39 % units better under the new equation than the old equation. For samples from the Sierra site, predictions of DOM by the new equation were 3.00% units more accurate than the original equation, although not all individual time periods were significant (Table 2.4).

DOM was consistently overpredicted with both the original and improved equation. The improved equation overpredicts DOM of samples from Petaluma by 0.74% units, and samples from the SFREC site by more than 1.67% units. The consistent overprediction of DOM indicates a systematic error either in the GAN Lab system or in the digestibility trial. However, because this over prediction was seen in the original equation as well as for CP, it is likely that the error lies within the GAN Lab NIRS system.

Overall, there was a greater improvement in prediction for CP than for DOM. Measures of *in vivo* DOM include inherent animal variation that is generally greater than the laboratory error associated with crude protein. On the other hand, laboratory CP error estimates were not available, so no statistical comparison of CP and DOM prediction improvement is possible.

#### Implications

The addition of California rangeland forage-fecal pairs made a significant improvement to the existing GAN Lab NIRS system. Further improvements are necessary, and could be made with additional digestibility trials on a wider variety of California rangelands. To increase usefulness of the system, these studies should include forages from the entire year rather than only the dry season.

Cattle producers should see an improvement in fecal NIRS predictions in early to mid 2004, when the GAN Lab will add the California data set, along with several other data sets from around the world, to the original equation. The GAN Lab has further plans to use locally weighted regressions to improve predictive ability of their equations. In this method, the computer obtains a spectral reading on a sample, which uses that reading to

identify the 100 most similar spectral samples in the database. These 100 samples are used to estimate parameters for an equation which predicts values for the specific unknown sample. Once available, this method is expected to yield much more accurate predictions (D. Tolleson, GAN Lab, Texas A&M University, TX, personal communication).

Scientific Name	Common Name	Petalum a May 14, 2003	SFREC May 30, 2003
		Percent of Cover	Forage
Arrana hanhada	Barbados Oat (Wild	9	
Avena barbada	Ual)		34
Avena fatua L.	Wild Oat		*
Brodiaea californica	California brodiaea		т
Bromus hordeaceus L.	Soft brome, soft chess		14
Bromus rigidus, roth	Ripgut Brome	14	*
Elymus caput medusae L.	Medusahead		19
Erodium cicutiaium	Filaree	*	*
Hordeum murinum L. ssp	Foxtail	11	
Hypochaeris glabra L	Smooth cat's-ear		*
Lolium multifloren	Annual rye	49	5
Lolium Perenne	Perinneal Rye	6	
Medicago polymorpha	Burr clover	*	
Stallaria media	Chicweed	*	
Trifolium hirtum	Rose clover		25
Trifolium subterraneum	Subclover	5	

Table 2.1: Species composition of Petaluma and SFREC rangeland sites.

\* Indicates minor species (constitutes <5% of forage cover)

	Date	Laboratory CP	In vivo DOM ± S.E.
la	01-Jul-02	5.2	53.96 ± 0.27
Ium	12-Aug-02	5.2	54.03 ± 1.04
eta	23-Sep-02	5.8	50.36 ± 1.82
م	04-Nov-02	6.0	50.55 ± 1.89
	01-Jun-02	4.3	56.17 ± 1.32
N N	23-Jul-02	4.1	53.01 ± 1.83
R.	03-Sep-02	4.5	52.95 ± 2.13
S	15-Oct-02	4.8	52.59 ± 3.52
	26-Nov-02	5.1	50.89 ± 0.53

**Table 2.2:** Laboratory CP and *in vivo* DOM values for Petaluma and SFREC digestibility trial forages.

		Predicted CP +/- SE		Predicted I	DOM +/- SE
Location	Date	Original Equation <sup>1</sup>	Improved Equation <sup>2</sup>	Original Equation <sup>1</sup>	Improved Equation <sup>2</sup>
3a	01-Jul-02	8.28 ± 0.33	6.62 ± 0.25	59.46 ± 0.26	58.49 ± 0.34
L III	12-Aug-02	7.12 ± 0.05	4.54 ± 0.28	56.54 ± 0.48	54.20 ± 0.15
eta	23-Sep-02	7.28 ± 0.29	$5.23 \pm 0.35$	56.74 ± 0.17	49.99 ± 0.18
L L	04-Nov-02	7.60 ± 0.25	5.78 ± 0.40	56.60 ± 0.29	49.19 ± 0.43
	01-Jun-02	7.89 ± 0.42	4.50 ± 0.19	57.33 ± 0.21	55.19 ± 0.42
U U	23-Jul-02	7.71 ± 0.37	4.03 ± 0.14	58.28 ± 0.20	56.09 ± 0.48
L R	03-Sep-02	9.22 ± 0.29	4.58 ± 0.34	58.00 ± 0.05	54.14 ± 0.14
L R	15-Oct-02	9.94 ± 0.17	4.94 ± 011	58.64 ± 0.13	54.91 ± 0.19
	26-Nov-02	9.28 ± 0.16	5.83 ± 0.43	56.71 ± 0.22	53.63 ± 0.31

Table 2.3: Means of DOM and	l CP predicti	ons by the original	and improved equations.
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 $<sup>^{1}</sup>$  Original equation represents predictions made with the original GAN Lab warm season equation <sup>2</sup> Improved equation represents predictions made with the improved GAN Lab warm

season equation
**Table 2.4:** LS means of DOM and CP error (predicted - laboratory/determined) predictions by the original and improved equations at Petaluma and SFREC rangeland sites.

		СР				DOM	
Location	Date	Original <sup>1</sup>	Improved <sup>2</sup>	P-value	Original <sup>1</sup>	Improved <sup>2</sup>	P-value
a	01-Jul-02	3.08	1.42	<0.01	5.5	4.53	0.64
Inn	12-Aug-02	1.92	-0.66	<0.01	2.52	0.18	0.26
eta	23-Sep-02	1.48	-0.58	<0.01	6.38	-0.37	<0.01
٩	04-Nov-02	1.60	-0.22	<0.01	6.14	-1.36	<0.01
	01-Jun-02	3.59	0.20	<0.01	1.17	-0.98	0.48
U U	23-Jul-02	3.61	-0.07	<0.01	5.27	3.08	0.47
R.	03-Sep-02	4.72	0.08	<0.01	5.04	1.19	0.21
S	15-Oct-02	5.14	0.14	<0.01	6.04	2.31	0.22
	26-Nov-02	4.18	0.73	<0.01	5.82	2.74	0.31



Figure 2.1: Predictions of CP with standard error at the Petaluma rangeland site.

Figure 2.2: Predictions of CP with standard error at the SFREC rangeland site.





Figure 2.3: Predictions of DOM with standard error at the Petaluma rangeland site.

Figure 2.4: Predictions of DOM with standard error at the SFREC rangeland site.







Figure 2.6: Differences between original and improved predictions and laboratory CP content for samples from the SFREC rangeland site.



Figure 2.7: Differences between original and improved predictions and laboratory DOM content for samples from the Petaluma rangeland site.



Figure 2.8: Differences between original and improved predictions and laboratory DOM content for samples from the SFREC rangeland site.



# Chapter III

# Demonstration of the Effect of Addition of California Data to the Texas A&M Grazinglands Animal Nutrition Laboratory Near-Infrared Reflectance Spectroscopy Equation at Five California Rangeland Sites

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ABSTRACT: The annual forage based rangelands of California, along with the state's Mediterranean climate, makes the beef cattle production system of California unique to the United Sates. Annual rangeland forages germinate with fall rains and grow through the end of the rainy season in the spring. When the spring rains cease, the annual grasses mature and senesce. The forage declines in quality due to increasing lignification. declining crude protein content and declining digestibility. This decrease in quality often requires cattle producers to provide supplements in order to maintain production, but it is difficult to know when and how much to supplement. The Grazinglands Animal Nutrition Laboratory (GAN Lab) provides commercial near-infrared reflectance spectroscopy (NIRS) of feces to predict CP and DOM of the consumed forage diet. Accurate predictions of unknown samples depend on the equation data set containing samples from the same population. The GAN Lab system includes samples from Texas, the Mid-west and the lower part of Western Canada. California producers have been using the system despite it never having been validated for California conditions. A series of digestibility trials were conducted to test the existing equation and to improve its predictive capability for California forages (Chapter II). In this study, forage and fecal samples were collected at five California range sites for 1 calendar year every 6 wks throughout the dry season and once monthly throughout the green season. Predictions of samples from Eureka did not differ by equation or season for DOM or CP. For valley and foothill sites, predictions of CP differed between the original and improved equation only during the dry season, while DOM differed by equation during the green season. Results demonstrate that the addition of California data to the Texas A&M NIRS dataset effect and improve prediction capability of the NIRS equation. Additional forage fecal pair data from a wider

variety of forage and season are needed to improve the predictive capability of the GAN Lab NIRS equation for California rangeland forages.

11.

#### Introduction

Beef cattle in California graze a unique rangeland composed mostly of non-native annual species which germinate with fall rains (Hart et al., 1932b; Pitt and Heady, 1978). The predominance of annual species, and the Mediterranean climate makes research and knowledge from other systems difficult to apply to California rangelands.

The pattern of annual forage growth in California was described by Bentley and Talbot in 1951 and divided into three seasons of availability for cattle: (a) the inadequate green season, (b) the adequate green season and; (c) the inadequate dry season. The inadequate green season begins with germination and extends to the beginning of warm spring weather. Although the nutritional quality of this new forage is high, both the height of forage and absolute amount available may limit voluntary intake by cattle. In addition, remaining dry forage from the previous year is of low nutritive value due to shatter and leaching of nutrients by rainfall. Warmer temperatures spur rapid spring growth, which characterizes the adequate green season. During this period, there is sufficient quantity and quality of forage to meet the nutritional demands of grazing beef cattle. The inadequate dry season begins when soil moisture declines to a point where the annual forages mature and senesce (George et al., 1985c; George et al., 1985a; George et al., 2001a; Young et al., 1973). With this maturation comes a decline in quality and palatability that prevents cattle from consuming enough forage to meet their nutritional needs. Throughout the dry season, forage quality continues to decline due to bleaching, shatter and decomposition, although the extent of the quality decline varies among forage species and location. Of particular concern for many cattle producers is the loss of crude protein (CP), which is necessary for growth, lactation and gestation in grazing cattle

(George et al., 2001b). Supplementation with forages, grains or minerals is often needed to maintain acceptable levels of production.

The varied topography, climates, soils and degree of human influence throughout California has resulted in a collection of highly diverse rangelands. This diversity is increased by natural yearly variation due to rainfall, temperatures, soil moisture, and other environmental factors (Hart et al., 1932a; Pitt and Heady, 1978). In addition, California's moderate climate allows invading species from around the world relatively easy colonization. Plant species have different nutritional values, and vary in both absolute quantity and accessibility to cattle throughout the year. In addition, the nutritional value varies within plant species according to stage of maturity.

For cattle producers, nutritional management is further complicated by selective grazing. Different classes of cattle graze differently and even within class, individual animals graze differently. On these highly diverse rangelands, predicting nutrient intake by any given group of cattle, and their resulting nutritional status, has been challenging and ultimately unsatisfactory.

The Grazinglands Animal Nutrition Laboratory, or GAN Lab, at Texas A&M University in College Station, Texas, uses near-infrared reflectance spectroscopy (NIRS) of fecal samples to predict the CP and digestible organic matter (DOM) of forages consumed by grazing livestock. In the study reported in Chapter II, the GAN Lab system was investigated for appropriateness for California rangelands. In conjunction with that study, forage and fecal samples from 8 California rangeland sites were collected to further characterize the rangelands and provide cattle producers with expected NIRS results for different rangeland forage types.

#### **Materials and Methods**

To better characterize California rangelands, and to give cattle producers a library of standard NIRS and nutrient values for rangeland forage, forage and fecal samples were collected from 5 rangeland sites throughout Northern California. Southern sites were eliminated due to drought conditions experienced in 2002.

Forage and fecal samples were collected on the same schedule as the harvests occurred at SFREC as described in chapter II. Briefly, samples were collected at 6 wk intervals throughout the dry season. Collections started the week of June 11 and continued through November 26, 2002. Starting in January, 2003, samples were collected the first week of each month until June, 2003. Samples were collected from the same pasture as cattle grazed, and so the collection site on each ranch moved with the cattle. In addition, samples were not collected when cattle were grazing irrigated pasture or public grazing lands.

At each site, range forage was collected by hand sampling. Care was taken to mirror the diet of the cattle by avoiding species known to be unpalatable at the given phenological stage and to collect species known to be grazed, either by observing the cattle or finding signs of grazing. Samples were dried at 50° for 72 hrs prior to grinding through a 1 mm screen on a Wiley Mill. Forage samples were analyzed by standard analysis for CP, acid detergent fiber, neutral detergent fiber and ash. Fecal samples were collected according to guidelines established by the GAN Lab at Texas A&M and were frozen after collection until sent by 2 d mail to the GAN Lab for NIRS analysis.

Method of determining species composition differed by site, but was limited to variations of the step-point method, where species are identified at specified intervals along a transect (CRRIC), and the double-sampling method (USDA). Forage production was determined by clipping at least 10 plots at each site.

# Statistical Analysis

NIRS data were separated into four groups; dry season data from the valley and foothill sites, green season data from the valley and foothill sites, dry season Eureka data, and green season Eureka data. The Eureka site has highest percent perennial forage, and as such, fits the original NIRS equation more closely than any of the other sites. The different species composition, climate, and season of growth at the Eureka site justifies analyzing this data separately from the other sites. The remaining sites, all located in the Central Valley and surrounding foothills, are similar in terms of species composition, percent annual, climate and growth curve. This data set, called the valley and foothill data, were separated into green and dry season data to analyze differences in predictive capability between these seasons. Also, the improved equation includes only samples from California's dry season and, as such, should not have a significant effect on predictive ability of green season samples.

Data were analyzed using the using the Proc GLM in SAS (Cary, NC) to determine the effect of season (dry and green) and forage type (valley and foothill vs. Eureka) on prediction by the original and improved equation.

# Site descriptions:

Sierra Foothills Research and Extension Center (SFREC): The SFREC is located 95 km northeast of Sacramento, and 24 km east of Marysville. Forage and fecal samples were collected throughout the facility, but cattle typically graze in oak grasslands. Average rainfall is 71 cm/year and the terrain ranges from rolling hills to steep hillsides.

**Eureka**: The Eureka site was located on a private ranch near the town of Kneeland, approximately 19 km southeast of Eureka. Forage and fecal samples were generally taken from a pasture with south-western exposure. Although some of the pasture is flat, it falls off steeply in many places. The area is known for high rainfall and frequent fog. It is located in one of the wettest areas of the state, and compared to the rest of the sites, has the highest percent of perennial forage species, and latest maturing, rangeland.

Madera: The Madera site was located on a private ranch about 19 km east of Madera and 24 km north of Fresno. Samples were collected from cattle grazing blue oak savanna. The terrain is moderately hilly with the ranch headquarters at 213 m above sea level. Average rainfall is approximately 40 cm/yr.

Yolo: Another private ranch was used for the Yolo site. It is located north of the town of Winters, approximately 48 km west of Sacramento on the west side of the Sacramento Valley. Samples were collected from cattle grazing in the lower foothills on oak studded annual grasslands. Average rainfall is 60 cm/year and the terrain ranges from flat to steep hillsides.

Lake Berryessa: This ranch is on the shores of Lake Berryessa in Napa County. The ranch is 65 km northeast of the city of Napa, and has a hilly terrain. In some places the terrain is quite steep, but is more gentle towards the base of the ranch. Cattle graze an oak grassland with some perennial forage species, particularly bunchgrasses. Average rainfall is about 67 cm/yr.

#### **Results and Discussion**

#### **Forage Production**

The date chosen to obtain species composition may not have corresponded with peak standing crop. Forage continued to grow later in 2003 than in normal years due to significant late spring rains with warm temperatures. In addition, these measurements of forage production represent the production of a single year rather than the long-term average, which may differ greatly from the value reported in Table 3.1. Forage production is provided to characterize each site and should not be used to compare sites.

Madera and SFREC had higher forage production than their respective historical averages. At SFREC, the historical average for May 1 from 1995 to present is approximately 3150 kg/ha, compared to approximately 4700 kg/ha in this study. Madera's average yearly forage production from 1939 to present is 2466 kg/hectare, compared to 4150 kg/ha in this study. (N. McDougald, Madera County Cooperative Extension, personal communication). The higher forage production in 2003 is most likely a result of a lengthened growing season due to late spring rains. Data for historical production is not available for the other sites.

## **Species Composition**

Species composition was obtained near the end of the year long sampling period (Table 3.2) to characterize each site. Ranking of species may differ considerably from the reported values if different rainfall patterns and quantities had occurred. Forage and fecal samples were collected over 2 forage years. The species composition reported reflects the second forage year and the species composition of the first forage year probably differed from the second.

Eureka had the highest percent of perennials in the sampling year at 19% of the forage cover. The contribution of perennials to the rangeland system suggests that Eureka fecal samples could be better predicted with the original GAN Lab equation than other sites because the original equation was almost completely based on fecal samples from perennial forages. Lake Berryessa had the second highest percent of perennials with 8% of forage cover. However, perennials were present in only one of the fields grazed during the study, and probably had a minor influence on predictions.

The species present at the valley and foothill sites are similar to those reported in the literature, particularly when major species are considered (Pitt and Heady, 1978). Fewer of the species reported by McNaughton in 1968 are the same, but this may be a result of the McNaughton study being isolated to one rangeland location or the fact that his rangeland had not been grazed for 5 yrs prior to determining its species composition. Some minor species listed in the older California reports were not found at our sites, but they may still be found on some California rangelands. The short duration of our study, the time of year sampling occurred, and the relatively small area sampled, all affect the species composition determined and could explain why some species found in earlier California literature were not found on our rangeland sites. Alternatively, the differences in species composition between this study and previous California literature may indicate a true shift in composition due to competition, management practices, yearly weather variation or invasion by foreign species that have occurred in the last 20 yrs. The composition of annuals on California rangelands does not appear to have changed in the

last 60 yrs. In 1942, annuals comprised 98% of the rangeland plant cover in the foothills of the San Joaquin Valley (Talbot and Biswell, 1942). In our study, annuals comprised 97.6% of the major species at the valley and foothill sites.

#### Laboratory Analysis

The CP, ADF, NDF, and ash values are reported on a DM basis for hand sampled forage over the year at the intervals described above (Table 3.3). Values from hand sampled rangelands represent, in general, nutrients available and could vary greatly from the actual diet of grazing cattle. Cattle are selective grazers and, as such, consume different forage species and parts of forage plants and in different ratios than are collected in hand samples, no matter how rigorously the hand samples were collected to measure what the cattle actually ate. Variations also exist between individual animals and classes of cattle. Collecting a hand sample that accurately reflects the nutrient intake of a herd is not likely, and there is no way of determining if, or when, the hand sampled forages are the same as the forages consumed. Hand sampling has historically been the best method available but, at best, it is a poor method to determine cattle diets in order to make nutritional management decisions.

Although hand sampled forages do not represent the nutrient value of the forage consumed among sites, the values can be used to characterize the forage growing at each site because samples were collected in the same method at each site throughout the year.

For all forage constituents tested, season was the only significant predictor (P<.01). Site and the site\*season interaction were not significant (P=.87, P=.68). The significant difference in laboratory values of the hand clipped forage samples between dry and green seasons indicates the need for forage-fecal pair data from both seasons in a

predictive equation. (Table 3.3) As expected, CP was highest during green up and rapid forage growth, and declined through the dry season (P<0.01).

# Near Infrared Reflectance Spectroscopy

Least square means of prediction differences (original equation – improved equation) for each site are in Table 3.4. The CP and DOM values predicted by the Texas A&M GAN Lab original equation and the improved equation, which includes samples obtained from California rangelands from Chapter 2, are listed in the appendix.

# **Crude Protein**

There was no equation or season effect on CP predictions for samples from the Eureka site (p=.63). This is not surprising as data added to create the improved equation did not include fecal samples from forages similar to the Eureka site.

The predictions for CP from the original and improved equations were significantly different for valley and foothill sites during the dry season (P<0.01), but not during the green season (P>0.01) (Table 3.4, 3.5) (Figure 3.1 - 3.5). The seasonal effect on differences between the original and improved predictions is likely due to the fact that only data from the dry season was included in the improved equation. With the addition of dry season California samples, it is reasonable that the improved equation gives more accurate predictions because the improved prediction equation includes similar data. Because no green season data was added, little change in predictive ability for that season occurred. Predictive ability for the dry and green seasons had opposite slopes due to the greater magnitude of change in predictive ability during the dry season, as evidenced by the least square means for CP.

# **Digestible Organic Matter**

DOM predictions for valley and foothill sites by the original and improved equation were significantly different during the green season (P<.01), but not during the dry season (P>0.01) (Table 3.5). New predictions of DOM were lower than the original predictions during the dry season and higher than original predictions in the green season. (Figure 3.6 - 3.10) Because DOM of dry, cured forages is lower than green forages, this decrease is logical when California's dry forages are compared to the perennials of other systems. No difference between predictions by equation or season was significant for the Eureka site.

#### Implications

The improved GAN Lab NIRS equation more accurately predicts CP of California's valley and foothill dry season forage, but green season predictions are not affected. This is important because that is the time of the year when CP is most limiting in the beef cattle production cycle. However, additional digestibility studies on a greater variety of California rangeland forages are necessary to further improve predictions. These studies should include at least a full year of forage-fecal pairs to account for the variation in forage quality between the dry and green seasons.

Although DOM predictions for the dry season on valley and foothill ranges are not different under the improved equation, green season predictions differ. Additional work in dry and green season DOM is necessary to determine whether DOM predictions are accurate.

Location	Date	DM Production <sup>3</sup> (kg/ha)
Eureka	June 10, 2003	3100
Lake Berryessa	April 16, 2003	2500
Madera	May 8, 2003	4150
SFREC	May 30, 2003	4700
Yolo	April 16, 2003	2200

Table 3.1: Dry matter forage production at five California rangeland sites

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<sup>&</sup>lt;sup>3</sup> Production rounded to the nearest 50 kg

<b>Table 3.2:</b>	Species composition	at 5 California	rangelands sites
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Scientific Name	Common Name	Eureka June 10, 2003	Lake Berryessa April 16, 2003	Madera May 8, 2003	SFREC May 24, June 4, 2003	Yolo April 26, 2003
			Pe	rcent of Forage C	over	
Agroseris heterophylla	Annual agroseris					*
Poa annua	Annual bluegrass	8				
Lolium multiflorum	Annual rye		19	*	5	
Aegilops triuncialis	Barbed goatgrass					*
Convolvulus L.	Bindweed		*			
Medicago lupulina	Black medic					*
Brassica nigra	Black mustard		*			
Erodium botrys	Broadleaf filaree			*		
Poa bolbosa	Bulbous bluegrass		*			
Medicago polymorpha	Burr clover					*
Brodiaea californica	California brodiaea				*	
Geranium carolinianum	Carolina geranium		*			11
Geranium mole	Dovesfoot geranium					*
Amsinckia menziesii	Fiddleneck		*	*		
Erodium cicutarium	Red-stem filaree		*	6	*	*
Hordeum murinum L. ssp leporinum	Foxtail		14			
Vulpia myuros	Foxtail, rat-tail or annual fescue	9	*	9		*
Stellaria media	Hairy chickweed			*		
Carduus pycnocephalus	Italian plumeless thistle					*
Hordeum geniculatum	Mediterranean barley			8		
Elymus caput medusae L.	Medusahead				19	
Dactylis glomerata L.	Orchard grass		8			
Plagiobothrys nothofulvus	Popcorn Flower			*		
Bromus rigidus, roth	Ripgut Brome			*	*	
Trifolium hirtum	Rose clover		*		25	10
Lolium perenne	Perennial Rye	11				
Hypochaeris glabra L.	Smooth cat's-ear			7	*	12
Bromus hordeaceus L.	Soft brome, soft chess	24	23	45	14	14
Lotus purshianus	Spanish clover			*		
Trifolium subterraneum	Subclover	23				
Anthoxanthum odoratum	Sweet vernalgrass	8				
Juncus bufonius L.	Toadrush			*		
Trifolium variegatum	Tomcat clover			*		
Eramucarpus setigerus	Turkey mullein		*	*		
Bromus alopecuros	Weedy brome					13
Festuca occidentalis	Western fescue		*			
Trifolium microcephalum	Whitetip Clover			*		
Avena fatua L.	Wild oat		21		34	14

\*indicates minor species providing less than 5% of cover

	Eu	reka	Lake Be	erryessa	Ma	dera	SF	REC	Y	olo		P-val	ues
	dry	green	dry	green	dry	green	dry	green	dry	green	Site	Season	Site*Season
% CP	7.10	12.52	4.05	14.87	4.78	11.67	4.48	11.68	4.63	14.78	0.87	<0.01	0.68
% ADF	44.13	38.57	47.60	35.42	47.50	40.05	47.85	41.65	47.93	38.32	0.79	<0.01	0.83
% NDF	70.93	61.70	70.32	51.50	66.03	54.03	67.98	57.18	66.47	53.82	0.52	<0.01	0.80
% Ash	4.65	6.68	7.60	11.13	6.78	12.17	6.80	9.88	8.17	10.87	0.02	<0.01	0.74

**Table 3.3:** Mean laboratory values of hand clipped<sup>4</sup> samples by forage season at five California rangeland sites.

<sup>&</sup>lt;sup>4</sup> Clipped to represent what cattle could have consumed

	Eur	reka	Lake B	erryessa	Ma	dera	SFF	REC	Y	olo
	CP	DOM	CP	DOM	CP	DOM	CP	DOM	CP	DOM
dry	0.72	-0.31	2.23	1.72	2.26	0.55	2.55	1.03	2.10	0.78
green	-0.32	0.30	0.01	-1.90	0.23	-2.57	-0.12	0.75	-0.93	-2.95
p-value	0.63	0.99	0.02	0.02	0.03	0.04	<0.01	0.89	<0.01	0.03

Table3.4: LS means of prediction differences (original equation - improved equation) by season for CP and DOM at five California rangeland sites

Eureka						Valley an	d Foothill	5
	CP	p-value	DOM	p-value	CP	p-value	DOM	p-value
Dry	0.72	0.14	-0.31	0.69	2.29	<0.01	1.02	0.12
Green	0.3	0.44	-0.32	0.61	-0.21	0.50	-1.67	<0.01

**Table 3.5:** Comparison and significance of LS means of prediction differences (original equation - improved equation) for CP  $\varepsilon$  DOM at Eureka and Valley-Foothill sites.

<sup>&</sup>lt;sup>5</sup> Valley and foothill sites include Lake Berryessa, Madera, SFREC and Yolo.



**Figure 3.1:** Comparisons of predictions of CP content of rangeland forage at Eureka rangeland site.

Figure 3.2: Comparisons of predictions of CP content of rangeland forage at the Madera rangeland site.





**Figure 3.3:** Comparisons of predictions of CP content of rangeland forage at the Lake Berryessa rangeland site.

Figure 3.4: Comparisons of predictions of CP content of rangeland forage at the SFREC rangeland site.







Figure 3.6: Comparisons of predictions of DOM of rangeland forages at the Eureka rangeland site.





Figure 3.7: Comparisons of predictions of DOM of rangeland forages at the Madera rangeland site.

Figure 3.8: Comparisons of predictions of DOM of rangeland forages at the Lake Berryessa rangeland site.





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Figure 3.9: Comparisons of predictions of DOM of rangeland forages at the SFREC rangeland site.

Figure 3.10: Comparisons of predictions of DOM of rangeland forages at the Yolo rangeland site.

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original equation

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Location	Date	Production (kg/ha)
Petaluma	April 14, 2003	2360
SFREC	May 30, 2003	4680

Appendix Table A: Forage Production at SFREC and Petaluma Rangeland Sites.

Appendix Table B: Laboratory values (%DM) for hand sampled rangeland	forages at
5 California sites	

	Date	СР	ADF	NDF	Ash
	07/31/02	5.7	40.1	67.5	5.7
	09/03/02	4.8	47.5	76.9	4.1
	10/14/02	4.4	52.2	79.3	3.0
a	12/03/02	5.3	47.1	73.6	3.9
sk	01/03/03	63	48.8	77.6	4.7
IL	02/03/03	13.3	40.4	63.9	9.3
E	03/04/04	10.6	37.3	61.9	6.3
	04/08/03	22.6	24 4	38.7	8.5
	04/30/03	17.0	33.4	54.5	7.4
	06/02/03	13.5	36.7	60.0	5.8
	07/25/03	4 2	45.9	71.6	6.9
-	09/05/02	3.0	46.2	74 4	6.6
222	10/28/02	4.2	48 7	65.3	6.7
<b>ye</b> :	12/05/02	8.6	46.2	66.7	7.1
E.	01/06/03	22.9	36.3	47 1	15.0
Sei	02/04/03	19.4	26.8	44 4	12.4
	02/04/03	15.7	27.9	40.8	12.1
Ike	01/02/02	15.7	33.5	48.8	11 3
Ľ	04/03/03	75	418	61.2	89
_	05/08/03	1.5	41.0	70.0	10.2
	07/02/02	4.0	47.8	68 1	73
	01/25/02	4.0	46.8	61.9	53
	09/05/02	4.2	40.0	68 5	6.8
~	10/15/02	5.0	40.0	65.4	11 4
ers	11/26/02	8.0 19.1	47.0	45.4	23.3
p	01/06/03	18.1	30.4	43.4	12.5
<b>U</b> a	02/01/03	12.2	40.2	53.4	10.6
	03/07/03	13.0	51.0 25.6	33.4 19 9	7 4
	04/03/03	8.8	33.0	40.0	7. <del>4</del> 9.1
	05/01/03	8.7	43.5	55.0	0.1
	06/05/03	5.3	47.4	60.8	7.7
	09/09/02	3.0	47.0	09.8	7.7
	10/16/02	5.1	48.8	00.2	7.7
	12/04/02	4.1	57.5	12.4	1.2
lo	01/06/03	16.5	39.4	54.0	11.3
Κo	02/07/03	22.1	24.5	40.0	13.4
	03/07/03	19.4	51.4	44.7	13.1
	04/01/03	16.2	57.9	50.4	10.9
	05/08/03	10.3	39.4	60.8	9.5
	06/05/03	7,2	47.4	63.4	8.9
	07/23/02	4.6	46.7	08.0	0.2
	09/03/02	3.8	46.1	05.0	0.4
	10/15/02	4.4	49.3	00.2	0.0
Ŋ	11/26/02	4.9	53.5	/1.2	7.4
S	01/07/03	14.2	42.0	57.2	9.7
G.	02/06/03	11.6	40.7	56.3	10.0
5	03/06/03	15.3	35.7	49.0	13.1
	04/01/03	13.2	38.6	52.6	10.7
	05/05/03	10.9	39.4	56.8	8.4
	06/04/03	5.1	49.3	72.1	8.6

		D	ОМ	C	P
Sample	Date	Original <sup>6</sup>	Improved <sup>7</sup>	<b>Original</b> <sup>6</sup>	Improved <sup>7</sup>
	07/31/02	59.90	60.32	10.65	10.22
	09/04/02	60.25	60.63	9.22	7.89
	10/14/02	58.85	56.85	8.76	7.98
_	12/03/02	61.57	60.40	9.68	10.76
eka	01/03/03	58.47	59.50	8.84	9.09
Eur	02/06/03	60.26	60.51	10.74	10.56
	03/04/03	60.13	59.05	10.97	10.14
	04/08/03	65.77	66.66	14.23	14.22
	04/30/03	63.99	65.99	13.20	11.12
	06/02/03	71.14	73.57	17.55	17.23

Appendix Table C.1: NIRS predictions of CP (% DM) and DOM (% DM) by original and improved GAN Lab equations for Eureka rangeland forages.

Appendix Table C.2: NIRS predictions of CP (%DM) and DOM (%DM) by original and improved GAN Lab equations for valley and foothill sites during dry season.

		D	OM	C	P
Sample	Date	Original 6	Improved 7	Original 6	Improved 7
Sa	07/25/02	58.91	55.08	8.04	4.87
ike /es:	09/05/02	61.23	58.81	9.19	5.89
La	10/18/02	57.08	56.34	8.20	5.09
2	06/06/03	58.74	58.86	7.19	7.85
	06/14/02	59.66	63.56	8.33	8.42
la	07/23/02	59.29	61.32	8.94	6.64
ade	09/11/02	60.24	56.25	7.88	6.29
Σ	10/15/02	60.35	56.77	10.42	7.13
	06/05/03	59.38	62.72	10.32	8.45
0	07/23/02	60.05	59.43	11.08	7.83
L L	09/03/02	58.84	58.23	8.14	5.31
SFF	10/15/02	60.52	56.47	11.74	8.62
	06/04/03	56.96	58.14	8.92	7.91
<u>0</u>	09/09/02	61.79	59.43	10.82	8.45
	10/16/02	60.22	57.97	10.13	7.11

<sup>6</sup> Original represents the predictions made by the original GAN Lab NIRS equation

<sup>7</sup> Improved represents the predictions made by the improved GAN Lab NIRS equation

06/05/03 58.50 60.78 11.24	10.32
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		DOM		CP	
Sample	Date	<b>Original</b> <sup>6</sup>	New <sup>7</sup>	<b>Original</b> <sup>6</sup>	New <sup>7</sup>
Lake Berryessa	12/05/02	59.69	60.99	12.21	9.20
	01/06/03	59.97	60.82	9.16	10.30
	02/04/03	62.42	65.60	13.35	13.19
	03/07/03	74.48	76.57	18.86	19.66
	04/03/03	68.12	69.51	16.71	17.90
	05/08/03	62.55	65.15	12.74	12.73
Madera	12/05/02	61.38	62.46	12.08	10.54
	01/02/03	60.71	63.96	11.44	11.47
	02/06/03	68.93	72.86	15.17	15.19
	03/07/03	70.91	73.31	18.61	17.72
	04/03/03	69.08	70.70	17.47	17.03
	05/01/03	62.01	65.16	11.10	12.57
SFREC	11/26/02	59.73	54.68	9.99	8.22
	01/07/03	60.43	56.78	8.97	10.24
	02/06/03	62.22	61.41	9.97	11.17
	03/06/03	61.57	58.75	9.84	9.98
	04/01/03	69.08	74.20	17.47	16.96
	05/05/03	64.74	67.44	15.03	15.44
Υοίο	12/04/02	60.36	59.62	11.54	9.28
	01/06/03	62.74	65.83	12.08	12.83
	02/07/03	70.69	75.15	15.56	19.22
	03/07/03	70.87	73.58	15.94	18.88
	04/01/03	67.58	70.50	16.13	17.23
	05/08/03	65.14	70.42	15.26	14.67

Appendix Table C.3: NIRS predictions of CP (%DM) and DOM (%DM) by original and improved GAN Lab equations for valley and foothill sites during green season.

Analysis of the Use of the Texas A&M Grazinglands Animal Nutrition Laboratory Fecal Near Infrared Reflectance Spectroscopy Prediction Equation with California Annual Rangeland Forages

**General Conclusions** 

Digestibility studies on dry season annual rangeland forages at two California rangeland sites were conducted to generate forage-fecal pair data to evaluate the predictive accuracy of the existing Texas A&M Grazinglands Animal Nutrition Laboratory (GAN Lab) warm season fecal NIRS equation. The addition of data from California rangeland forages to the original GAN lab warm-season equation improved (P<0.01) prediction of CP by 2 to 4% units. Addition of forage-fecal pair data from California rangelands also improved (P<0.01) predictions of DOM by 3 to 4% units.

In a separate study, fecal samples were collected from cattle grazing at five California rangeland sites for 1 calendar year. Forage samples were collected by hand clipping from the same pastures in which the fecal samples were collected. Fecal NIRS predictions of forage CP and DOM were obtained before and after addition of the California forage-fecal pairs to the original warm season equation. No difference in predictions of CP or DOM occurred between equations for samples from the Eureka site. This was not surprising because samples from the Eureka site were the most similar to the samples used to construct the GAN lab equations. Predictions of CP at the valley and foothill sites differed (P<0.01) between equations, with the original equation predicting higher CP content, during the dry season but not during the green season. This is consistent with the addition of forage-fecal data for the dry season and not the green season. In contrast, predictions of DOM at the same site differed (P < 0.01) between equations during the green season but not the dry season. The original equation predicted lower DOM for the green season than the original equation. However, reasons for the recorded improvement in DOM for the green season, from which no forage-fecal pairs were added, but not for the dry season, when they were added, are not evident.

Improvement in prediction accuracy was better in the first study than in the second probably because, the samples that were analyzed in the first study were the same samples that had been added to the equation. The system would "recognize" those samples and predict them more closely than samples that did not have corresponding samples in the database. Smaller improvements were seen in the second study and, although the differences between the predictions of the two equations were not always significant, the improved NIRS equation has greater predictive power for fecal samples from cattle grazing California rangeland forages because the new equation can "recognize" the chemical bonds present in California samples.

The observed improvement in prediction of CP and DOM by the improved equation has important implications for California cattle producers, especially those whose cattle graze valley and foothill rangelands, as it will allow more exact nutrient and supplement management, which reduces feed costs, improves cattle performance, and minimizes environmental impact.

California cattle producers should see an improvement in fecal NIRS predictions in early to mid 2004, when the GAN Lab will add the California dataset, along with several other data sets from around the world, to the original equation. The GAN Lab also has plans to use locally weighted regressions to improved predictive ability of their equations. Once available, this method is expected to yield much more accurate predictions of CP and DOM content of rangeland forages consumed by grazing cattle around the world.

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