GLOBAL LIVESTOCK CRSP

Development Of Portable NIRS Technology To Monitor Grazing Animal Nutrition In Mongolia

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Understanding nutritional status of grazing animals is a critical step in improving livestock productivity in Mongolia. Utilization of near infrared reflectance spectroscopy (NIRS) on feces provides adequate assessment of diet quality, yet initially requires substantial laboratory infrastructure and capacity. The application of portable NIRS in remote settings such as those found in Mongolia shows promise as both a research and management tool, as it allows for rapid assessment of diet quality in the field. Until Mongolian specific calibrations are developed, application of fecal NIRS there is dependent on similarities in fecal chemistry between native and US animals. Fecal chemistry of Mongolian cattle and yak were highly analogous to US cattle, whereas Mongolian sheep and goats exhibited greater differences in fecal chemistry as compared to their US counterparts than did the larger ruminants. Due to these similarities, diet quality from Mongolian ruminant fecal samples can be determined using current US calibrations until either Mongolian specific or Mongolian enhanced calibrations are developed. Fecal NIRS can also be used to discriminate between pairs of sympatric species (i.e. cattle and yak, sheep and goat, horse and khulan). Further development of this discriminant technique will facilitate more efficient collection of samples (by non-livestock oriented individuals), thus expediting nutritional monitoring in large landscapes. Timely assessment of diet quality via fecal NIRS will augment the forage quantity measurements provided by the Gobi Forage project and provide a two-pronged decision support package for pastoralists and agriculture professionals. Portable NIRS yielded results comparable to static NIRS and can thus be used in field conditions. This "take the lab to the sample" approach with portable NIRS can make near-real time, location specific nutritional monitoring available to the herders of Mongolia's vast grazinglands.

Background

Nutritional status of grazing animals results from a combination of forage quantity and quality. NIRS of feces is a non-invasive method by which diet quality can be determined. Fecal NIRS has been employed to monitor the nutritional status of grazing animals in the US and subsequently either extended to or developed independently in other countries. This work was part of the GL-CRSP Livestock Early Warning System (LEWS) project in East Africa. Previous fecal NIRS labs have employed the central lab concept using bench top, static NIRS equipment, i.e. after the US model. In countries with poor travel infrastructure and or mail capabilities, however, the central lab may not always be utilized as effectively for diet quality to be ascertained in a timely fashion. For this reason, a new concept is being applied in Mongolia, namely a "take the lab to the sample" strategy. The recent development and improvement of portable NIRS instruments has provided for the possibility of highly mobile laboratories. The objective of fecal NIRS work in Mongolia is to determine the ability of portable NIRS to monitor diet quality of grazing livestock in remote conditions.

Several questions exist as to the feasibility of applying portable NIRS to fecal samples for determining diet quality of grazing animals in Mongolia.

1. *How well does the portable equipment duplicate the performance of bench top equipment?*
2. *Is the fecal chemistry of Mongolian livestock similar to US livestock?*
3. *Can fecal NIRS calibration equations be developed on portable equipment to predict diet quality of Mongolian livestock?*
4. *What are the logistics associated with delivering portable NIRS to herders in the Mongolian countryside?*

Research projects have been designed to answer these questions. Preliminary results of that research are presented here.

Preliminary Findings

Question 1. How well does the portable equipment duplicate the performance of bench top equipment? Prior to a full scale effort to deploy portable NIRS in Mongolia, the first task was to determine the ability of portable NIRS to predict animal diet quality from fecal samples. The NIR spectrometer is housed at the Grazingland Animal Nutrition (GAN) Lab at Texas A&M University and

Figure 1. Portable NIRS set up for acquiring spectra in the field with hand held fiber optic probe. High Mountain Research Station, Mongolia. Photo by Doug Tolleson

diet quality calibrations developed on this equipment, serve as the "master" system for the LEWS projects. The initial step, prior to even purchasing a portable unit, was to determine the performance of static spectrometer derived calibrations when the spectral range was abbreviated to that of the portable unit. Predictive equations for dietary crude protein (CP) and digestible organic matter (DOM) using the fecal calibration sets for US cattle at 800 to 1700 nm resulted in RSQ and SE calibration (SEC) of 0.81;1.01 and 0.70;1.20 for percent CP and DOM respectively. Once the portable machine was obtained, the first task was to re-create a calibration originally developed on the static machine using spectra collected on the portable.

Near infrared absorbance spectra were collected on the portable spectrometer from a set of 42 sheep fecal samples. Diet reference chemistry values for CP and DOM from the previous study were matched with new spectra obtained on the portable spectrometer. Absorbance spectra from the portable spectrometer are visibly similar to those from the bench-top spectrometer. Predictive equation performance statistics (RSQ;SEC) were: 0.96;0.97, 0.92;0.84 and 0.32;12.3 for CP, DOM and random numbers respectively. Calibration results for the original static NIRS for this set were: 0.95;1.08 and 0.80;1.51 for CP and DOM respectively. Protein results were very similar between the two systems while the portable calibration DOM results were better than the static NIRS calibration.

Question 2. Is fecal chemistry of Mongolian livestock similar to US livestock? The next task was to evaluate the similarity of fecal chemistry between US and Mongolian livestock. Our hypothesis was that there would be enough similarity between the two such that whole scale development of Mongolian specific calibrations would not be necessary. In this case, Mongolian diet reference chemistry:fecal spectrum (D:F) pairs would serve as validations of the US equation performance and as enhancements to the existing calibrations. Fecal samples from cattle, yak, sheep, goats, horses, and khulan or wild ass *(Equus hemionus)* were collected in the Mongolian countryside in 2004 and 2005. These were collected either opportunistically during forage transect work, or specifically during NIRS training excursions. One indicator of spectral relatedness is Mahalanobis distance, basically a multivariate standard deviation. When new samples are evaluated against an existing calibration set spectral population, the Mahalanobis distance of each sample gives an indication of how closely related that sample is to the centroid of the calibration set. The smaller the value, the more spectral similarity to that population. In fecal NIRS calibrations, a value of greater than 8.0 is considered an outlier. US derived calibrations for cattle, sheep, goats and donkeys were used to determine Mahalanobis distance values for each corresponding species. Additionally, yak samples were predicted with the cattle equation and horse samples with the donkey equation. Mean ± SE Mahalanobis distance values for cattle, yak, sheep, goat, horse and khulan were 1.75 ± 0.21, 1.56 ± 0.11, 5.45 ± 1.33, 4.60 ± 0.61, 9.30 ± 0.78, and 13.6 ± 1.34 respectively. Cattle and yak values were well below the outlier determination. Mahalanobis distance values for sheep and goat were greater than the larger ruminants but still within an acceptable range. Fecal chemistry of the equine species, as indicated by NIR spectra, was very different between US and Mongolian animals. These values were well out of the acceptable range. Diet quality from Mongolian ruminant fecal samples can be determined using current US calibrations until either Mongolian specific or Mongolian enhanced calibrations are developed. Current US equine calibrations are not yet sufficiently robust for deployment in Mongolia.

The application of fecal NIRS in extensive landscapes such as those found in Mongolia will depend not only on scientific factors but on logistic ones as well. One difficulty encountered during fecal sampling, specifically by forage transect crews, has been positive identification of the livestock species being sampled. This can be especially difficult within multi-species herds. Sheep and goats for example produce fecal pellets which have a similar appearance. Close observation to visually observe the animal defecating is doable but time consuming. Opportunistic collection would be a more efficient use of time for these crews. An experiment was conducted in Mongolia to determine the ability of fecal NIRS to discriminate between three pairs of livestock or wildlife with similar fecal morphology. These pairs consisted of cattle and yak, sheep and goats, and lastly, horses and khulan. Fecal samples were collected with positive visual species identification throughout the growing season across diverse landscapes in Mongolia in 2004 and 2005. The large ruminant calibration resulted in an RSQ = 0.88 and SE = 0.16. Prediction of species

identity for 16 samples not included in the calibration set yielded 100% correct determinations. Similar values for the small ruminants were 0.66 and 0.28 respectively with 16 out of 20 correct identifications. Corresponding values for the equines, were 0.98 and 0.06 respectively with 12 out of 12 independent validation samples correctly identified. Fecal NIRS can thus be used to discriminate between these pairs of sympatric species. Development of this technique will facilitate more efficient collection of samples (by non-livestock oriented individuals) for nutritional monitoring purposes in large landscapes.

Question 3. Can fecal NIRS calibration equations be developed on portable equipment to predict diet quality of Mongolian livestock? This current project in Mongolia represents the first attempt at developing fecal NIRS diet quality calibrations on portable NIRS equipment. The first step in this process was to conduct small feeding trials to create Mongolian D:F pairs. The trial described here was conducted at the Research Institute for Animal Husbandry (RIAH) headquarters in Ulaan Baatar. Ten mature sheep (40.0 ± 5.0 kg) obtained locally were fed diets composed of Mongolian forages in 2x2 m pens with ad libitum water. Each trial lasted 7 days. Prior to being fed experimental diets, an adaptation trial was conducted in which all animals received the same diet. Afterward, each animal received a unique diet. Each animal took part in more than one feeding trial. Diets were offered at approximately 2% of body weight split into morning and evening feedings. Diet samples, refusals and fecal samples were collected on days 6 and 7 of each trial. Reference data were paired with spectra and used in calibration model development. A total of 63 D:F pairs were created. Due to logistic constraints on applying calibration models with current software for the portable unit, stepwise regression was used to develop diet quality predictive equations. At the time of this writing, DOM was still being conducted on these samples, so only CP will be reported here. Preliminary equation results for diet CP in Mongolian sheep were RSQ = 0.81 and SEC = 1.30. Threshold evaluation criteria for excellent fecal NIRS equations to determine CP are typically RSQ > 0.90 and SEC no greater than 2X the lab error (~ 0.50 in this instance) for the reference method. The values obtained in this calibration set are thus satisfactory, but could be improved. As further feeding trial data are obtained and more refined calibration techniques are applied, it is expected that these results will improve.

Table 1. Portable NIRS predicted values for fecal nitrogen (FN) in sheep and goats.

Sample ID	Date	Species	Herder	Aimag	FN
UGgt.0000001	May 17 2007	Goat	A	Umno Gobi	1.85
UGgt.0000002	May 17 2007	Goat	A	Umno Gobi	1.81
UGgt.0000003	May 17 2007	Goat	A	Umno Gobi	1.89
UGgt.0000004	May 17 2007	Goat	B	Umno Gobi	1.64
UGgt.0000005	May 17 2007	Goat	B	Umno Gobi	1.28
UGgt.0000006	May 17 2007	Goat	B	Umno Gobi	1.64
UGgt.0000007	May 17 2007	Goat	C	Umno Gobi	1.47
UGgt.0000008	May 17 2007	Goat	C	Umno Gobi	1.63
UGgt.0000009	May 17 2007	Goat	C	Umno Gobi	1.55
UGgt.0000010	May 17 2007	Goat	C	Umno Gobi	1.99
UGsh.0000001	May 17 2007	Sheep	A	Umno Gobi	1.53
UGsh.0000002	May 17 2007	Sheep	A	Umno Gobi	1.47
UGsh.0000003	May 17 2007	Sheep	A	Umno Gobi	18.7
UGsh.0000004	May 17 2007	Sheep	B	Umno Gobi	1.81
UGsh.0000005	May 17 2007	Sheep	B	Umno Gobi	1.73
UGsh.0000006	May 17 2007	Sheep	B	Umno Gobi	1.53
UGsh.0000007	May 17 2007	Sheep	C	Umno Gobi	1.82
UGsh.0000008	May 17 2007	Sheep	C	Umno Gobi	1.73
UGsh.0000009	May 17 2007	Sheep	C	Umno Gobi	1.63
UGsh.0000010	May 17 2007	Sheep	C	Umno Gobi	1.58

Question 4. What are the logistics associated with delivering portable NIRS to herders in the Mongolian countryside? The final step in our initial project to determine the feasibility of applying portable NIRS to monitor grazing animal nutrition in Mongolia was to identify and overcome the practical logistical problems one might encounter. Bench-top NIRS is performed under controlled conditions with respect to temperature, light and humidity. There are of course certain environments in which this is more difficult than others, even in a lab setting. In addition to the above mentioned factors, consistent sample presentation could be a problem with the portable equipment. The unit employed in this project utilizes a fiber optic probe which can be handheld (Figure 1) or mounted in a holding apparatus. Our primary goal was to re-create, as closely as possible, the bench-top NIRS laboratory configuration with portable equipment, thus developing and extending new technology.

Initially, only the NIRS equipment itself was moved from place to place. The drying and grinding equipment was established at three RIAH locations: 1) headquarters in Ulaan Baatar, 2) the High Mountain Research Station in Ikhtamir, and 3) the Gobi Research Station in Bulgan. With this suite of test locations established, a route will be evaluated in which the portable NIRS lab will arrive at each facility on a certain day each time period (monthly perhaps).

Fecal samples will be collected from herds in the area on day 1, dried over night, then ground, re-dried and scanned on day 2. Results will then be posted by various means such as print, radio or internet. The portable NIRS equipment has been used in training settings during field trips in 2005 and 2006, however on these occasions, only spectra could be obtained. No calibration equations had been developed nor was there software available to apply equations. In 2007 the combination of appropriate software and a completely Mongolian derived multi-species equation for fecal N allowed the first "road test" of the portable fecal NIRS lab. Twenty fecal samples, 10 each from sheep and goats were collected in Umno Gobi aimag within a 20 km radius of Bulgan. These were dried, ground, scanned and predicted under the conditions encountered on the trip. Notably, electricity was not available in Bulgan at the time with the exception of ~ 8pm to midnight. After several attempts, a portable generator strong enough to power the oven and grinder was located and pressed into service. Fecal N predictions for sheep and goats are presented in Table 1.

Practical Implications

Portable NIRS as applied in this project has several advantages and disadvantages that will affect its practical implementation. On the positive side, portable NIRS equipment is less costly than static systems and can be transported to remote locations for near real-time determinations of grazing animal diet quality. NIRS is a non-invasive and adaptable quantification technique, thus can be applied to a great many substances, not just animal feces. For instance, the technique has been used to sort mohair, from live animals prior to shearing, into fiber diameter groups. Disadvantages of the current configuration primarily involve ensuring adequate electrical power in remote locations to operate the associated drying and grinding equipment. Additionally, the portable NIR spectrometer utilized does not include software capable of developing predictive calibrations. This results in additional software and statistical training for NIRS technicians or creates the need for collaboration with other personnel to obtain the statistical expertise. The full exploitation of NIRS in Mongolia will require establishment of a central static NIRS unit in conjunction with functional wet chemistry laboratories in addition to the portable units. This will allow for both a stable ongoing calibration development environment and a highly mobile, in field, extension of these calibrations. Thus equipped, the Mongolian agricultural scientific community will be able to not only evaluate forages and feedstuffs in an academic setting, but in the realm of grazing animal production as well. By providing samples and feedback on the effectiveness of the portable NIRS technique, herders will be both contributors to and beneficiaries of this new technology.

Further Reading

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The Global Livestock CRSP is comprised of multidisciplinary, collaborative projects focused on human nutrition, economic growth, environment and policy related to animal agriculture and linked by a global theme of risk in a changing environment. The program is active in West and East Africa, Central Asia and Latin America.

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