# NEAR INFRA-RED MEASUREMENT OF NONSTRUCTURAL CARBOHYDRATES IN

#### **ALFALFA HAY**

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## Abstract

Recently documented benefits from afternoon versus morning cut forage have encouraged laboratory reporting of total nonstructural carbohydrate (TNC) values as part of forage quality testing. Our objective was to determine if infra-red spectroscopy (NIRS), which is being used in many forage testing labs, could be reliably used to quantify forage sugars in hay samples. We used two alfalfa (*Medicago sativa* L.) sample populations that were analyzed by wet chemistry for sugars and scanned by NIRS. The first set consisted of field-dried hay samples that were oven dried at 70°C and the second consisted of fresh, freeze-dried samples. TNC values were determined more precisely with NIRS than by wet chemistry.

**Keywords:** NIRS, alfalfa, forage quality, diurnal quality changes, total nonstructural carbohydrates.

#### Introduction

Cattle, sheep, and goats prefer afternoon-cut hay (PM) to that cut in the morning (AM) (Fisher et al. 1998, 1999). Yet these hays may have similar fiber and protein values but have a greater concentration of TNC in the PM-cut forage. Expected animal preference and production responses to feeding of PM-cut hay necessitates reporting TNC values. Forage quality tests are routinely based on NIRS and constituents are calculated from NIR spectra. The objective of this study was to test the ability of NIRS to predict TNC in alfalfa hay, especially given present instrumentation and software.

### **Material and Methods**

Hay Samples. Alfalfa, grown with furrow irrigation in semi-arid (270 mm precipitation), south central Idaho, and was cut with a sickle-bar mower at late bud stage (Kalu-Fick stage 4) during July, August, and September. The hay was field dried, baled, transported to Raleigh, North Carolina where it was fed in a preference trial (Fisher, et al 1998). Subsamples of hay were taken throughout the study, dried at 75°C in forced-draft oven, and ground successively through a Wiley shear mill and a Cyclotec abrasion mill to pass a 1-mm screen. Total nonstructural carbohydrates (TNC) were analyzed by a modified method described by Fisher and Burns (1987). Samples were scanned via Perstorp Analytical (Silver Spring, MD) Model no. 5000 near infra-red spectrometer using ISI version 4.01 software. Data were processed using modified partial least squares (MPLS) as described in the operating manual. The MPLS is often more stable and accurate than the standard PLS algorithm. Composition values are reported on a dry mater basis and TNC is the sum of each of the analyzed sugars.

Fresh Grab Samples. Alfalfa grab samples were taken at 3 h intervals during the 72 to 96-h period preceding forage cutting. Fresh samples were immediately placed on dry-ice, frozen, freeze dried, ground, and analyzed like the hay samples.

#### **Results and Discussion**

The hay and fresh grab samples were from the same field, except the hay samples were taken 3 to 6 months later while the hay was being fed (Table 1). The first obvious difference is that the drydown time from cutting to baling to feeding reduces the concentration of individual and total sugars and increases fiber components. With exception of the short chain polysaccharide fraction, other sugars and fiber fractions where measured with high precision (Table 1) in both hay and grab samples. Table 2 contains statistics on sugar, crude protein and fiber components of alfalfa hay samples cut at sundown and sunup on three dates. The correlations are less on these field samples (Table 2) because of limited variation in the data set. We are encouraged that sugars, especially TNC, are measured with sufficient precision that NIRS can successfully assess the readily soluble energy component contained in PM- and AM-cut alfalfa. This conclusion is strengthened by the excellent correlations obtained by Albrecht et al (1987) using broad based population of alfalfa breeder materials. The new software now available will enhance the opportunities to predict sugar levels in alfalfa hay samples using NIRS.

#### References

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**Table 1 -** Near infrared statistics as generated with modified partial least squares (MPLS) for sugars and fibers in freeze-dried and field-dried alfalfa.

Variable	N	Mean	SEC <sup>1</sup>	SEC	SECV <sup>3</sup>	SECV	MT <sup>5</sup>
				$RSQ^2$		$RSQ^4$	
Freeze Dried		g kg <sup>-1</sup>	g kg <sup>-1</sup>		g kg <sup>-1</sup>		
Monosaccharide	154	13.7	2.05	0.79	2.41	0.71	2,10,10,1
Disaccharide	152	25.5	2.72	0.85	3.34	0.77	4,10,10,1
SCP <sup>6</sup>	151	6.1	1.22	0.40	1.41	0.20	1,4,4,1
Starch	150	18.2	1.69	0.98	2.19	0.97	2,10,10,1
TNC	151	63.8	4.19	0.95	4.76	0.93	4,10,10,1
ADF	153	253.	3.7	0.99	4.1	0.98	1,4,4,1
NDF	151	323.	4.5	0.99	4.8	0.99	2,10,10,1
Field Dried							
Monosaccharide	80	11.9	1.03	0.92	1.44	0.84	1,4,4,1
Disaccharide	80	17.1	2.18	0.95	2.68	0.92	4,10,10,1
$SCP^6$	74	9.4	1.93	0.24	2.07	0.16	4,10,10,1
Starch	82	6.8	1.66	0.57	2.22	0.23	3,10,10,1
TNC	80	45.3	3.75	0.92	4.00	0.91	3,10,10,1
ADF	157	350.	6.5	0.99	7.7	0.99	2,10,10,1
NDF	157	464.	6.5	0.99	8.2	0.99	4,10,10,1

<sup>&</sup>lt;sup>1</sup>SEC = Standard error of calibration

<sup>&</sup>lt;sup>2</sup>SEC RSQ = Fraction of explained variance for SEC

<sup>&</sup>lt;sup>3</sup>SECV = Standard error of cross validation

<sup>&</sup>lt;sup>4</sup>SECV RSQ = Fraction of explained variance for SECV

 $<sup>{}^{5}</sup>MT = Math treatment$ 

<sup>&</sup>lt;sup>6</sup>SCP = Short chain polysaccharides

**Table 2** - Means, root mean square error (RMSE), and correlations (R-SQ) for sugars, crude protein, and forage quality characteristics in field-dried alfalfa hay samples as determined by NIR and laboratory wet chemistry.

	NIR			LAB			
	Mean	RMSE	R-SQ	Mean	RMSE	R-SQ	
	g kg <sup>-1</sup>	-	-	g kg <sup>-1</sup>	-	-	
Sugars <sup>1</sup>							
Monosaccharide	12.0	1.7	0.84	11.6	2.2	0.78	
Disaccharide	17.3	2.6	0.95	17.1	4.0	0.88	
$SCP^4$	9.4	0.4	0.93	9.0	2.4	0.47	
Starch	6.7	1.8	0.43	6.8	2.5	0.36	
TNC	45.0	3.8	0.94	44.0	6.9	0.85	
Crude Protein <sup>2</sup>	196	33	0.52	195	34	0.51	
Forage Quality <sup>3</sup>							
NDF	464	65	0.51	464	65	0.52	
ADF	350	56	0.48	349	58	0.47	
Cellulose	269	45	0.48	269	45	0.48	
IVTD	728	50	0.56	727	54	0.27	
Lignin	76	33	0.52	76	34	0.51	

 $^{1}$ n = 83,  $^{2}$  n= 78, and  $^{3}$ n = 162.  $^{4}$ SCP = Short chain polysaccarides.