Chapter 8.
Seasonal Variation of Hydrolyzable Carbohydrates in Pasture: A Comparison of Direct and Indirect Methods of Determination

ABSTRACT

Seasonal variation of pasture may include large changes in the concentrations of hydrolyzable carbohydrates and fibers. Dietary carbohydrates may be hydrolyzed or fermented in the horse. Accurate analysis of carbohydrate fractions in pasture is important for the evaluation of the diet of grazing animals, but laboratory methods traditionally calculate non-structural carbohydrate by difference. The objective was to examine seasonal variation in pasture and compare hydrolyzable carbohydrate concentrations in pasture, hay and supplements as analyzed by direct and indirect methods. One hundred seven pasture and hay samples and twenty-five samples of research supplements were collected, dried, ground and analyzed by difference for non-structural carbohydrate, directly for hydrolyzable carbohydrate. Non-structural carbohydrate as calculated by difference overestimated hydrolyzable carbohydrate in pasture, hay and supplement samples. Hydrolyzable carbohydrate was predicted from non-structural carbohydrate (r = .989; *P* = .0001).

Key Words: Hydrolyzable Carbohydrate, Non-structural Carbohydrate, Pasture, Season

Introduction

Accurate, precise and convenient analysis of carbohydrate fractions in pasture plants is important for evaluating the diet of pasture grazing animals. Physiologically, in the horse and other hind gut fermenting animals, carbohydrates may be hydrolyzed or fermented. Hydrolysis of carbohydrate yields glucose and other hexoses. In contrast, fermentation products are mainly acetate, propionate and butyrate. Metabolic efficiency is greater for glucose than for acetate (Blaxter, 1989), therefore division of carbohydrates by analysis should be into hydrolyzable and fermentable fractions.

Official methods for the Dairy Herd Improvement Association (DHIA) measure plant cell walls as neutral detergent fiber, NDF (Van Soest, 1991). Other laboratory methods, used by human nutritionists, measure total dietary fiber (TDF), which includes NDF plus soluble fibers, which are fermented (Hall, 1989). Non-structural carbohydrate (NSC), the portion found within plant cells, is estimated by difference. Ruminant nutritionists estimate NSC = 100 – (water + protein + fat + ash + NDF), but human nutritionists estimate NSC = 100 – (water + protein + fat + ash + TDF). Both estimates of NSC include some fermentable carbohydrate and therefore overestimate the hydrolyzable fraction.

Hydrolyzable carbohydrate may be measured directly (Davis, 1976; Smith, 1981). Referred to as total nonstructural carbohydrate (TNC), this analysis yields only the hydrolyzable fraction. Thus, TNC, as defined by the method, is synonymous with hydrolyzable carbohydrate (CHO-H). Our objective was to examine seasonal variation and compare pasture, hay and supplement concentrations of NSC, as estimated by difference using NDF, with CHO-H, measured by TNC analysis.

Materials and Methods

One hundred seven pasture and hay samples were collected, from October, 1995, to November, 1996. Of these, 83 were fresh samples collected from research pastures at the Virginia Tech MARE Center. Most of the pastures and hays were of a grass legume mix, many consisting of bluegrass and white clover. Pasture samples were collected by random clippings gathered from all sections of the pasture, and hay samples were collected using a core sampler and several bales selected randomly from each field and cut. Twenty-five samples of research supplements (10 from the sugar and starch supplement, 15 from the fat and fiber supplement) were also collected.

The samples were weighed, then dried for 24 h at 100º C, and dry matter was calculated. Dry samples were first coarsely ground and then ground again using a cyclone mill and a 1 mm screen. A subsample of approximately two grams was scanned through a Near Infrared Reflectance Spectrophotometer in order to generate a library of forage samples for future development of calibration equations. Each subsample was returned to its respective container. The samples were stored in sealed plastic containers placed inside a dessicator pending analysis. Approximately one gram of each sample was reserved for direct analysis of hydrolyzable carbohydrate; the remainder was sent to the Northeast DHIA Forage Laboratory

(Ithaca, NY) for proximate analysis. Total nonstructural carbohydrate (hydrolyzable carbohydrate) was determined using a direct analysis method (Smith, 1981).

Seasonal variation in CHO-H and NSC was summarized using means and standard errors. Data were analyzed using analysis of variance (SAS, 1988) to compare seasonal differences, and regression equations were fit using a graphics program (SlideWrite, 1995) to compare direct and indirect analysis data. When regression equations indicated results that were technically impossible (i.e. positive CHO-H when NSC = 0), the equation was forced through zero.

Results

In MARE Center pasture, seasonal variation was evident for hydrolyzable carbohydrate (CHO-H) and non-structural carbohydrate (NSC) "by difference" (Figure 8.1a). Non-structural carbohydrate "by difference" over-estimated CHO-H at all sample times except for November pasture (Figure 8.1b). The overestimation increased as CHO-H increased. Seasonal variation in CHO-H showed smooth increases and decreases, peaking in March and April. In comparison, NSC varied more randomly, perhaps due to differences in non-hydrolyzable components included in the estimation.

Direct analysis of CHO-H data were plotted against NSC (Figures 8.2, 8.3, 8.4 and 8.5). Linear regression estimated CHO-H from NSC using data from the sugar and starch supplement (Figure 8.2a, r = .683, P = .027), the fat and fiber supplement (Figure 8.2b, r = .775, P = .001), and the 107 pasture and hay samples (Figure 8.3, r = .707, P = .0001). Estimation of CHO-H from NSC using linear regression improved with combined data from the sugar and starch, and fat and fiber supplements (Figure 8.4a, r = .991, *P* = .0001). Data from the fat and fiber supplement were more similar to the pasture data than data from the sugar and starch supplement. Combined data from the fat and fiber supplement and the pasture provided an estimation of CHO-H from NSC using a quadratic equation (Figure 8.4b, r = .845, *P* = .00001). A combination of all data from both supplements and the pasture provided what may be the best overall picture (Figure 8.5). A quadratic equation, CHO-H = .154*NSC + .0136*NSC*NSC, provided the best fit (r = .989, *P* = .00001).

Discussion

As cool season grasses grow from leafy to stemmy stages, dry matter yields increase with increases in fiber and lignin, and protein and nonstructural carbohydrate concentration decrease (Blaser et al., 1986). These differences correspond with season, with rapid new growth occuring in the spring or during periods after rain, and maturation throughout the summer. As the leafy portion is grazed or cut, regrowth provides additional leafy material until the growing season ends. The increase in CHO-H and NSC found in the spring pasture sampled in this study agrees with data from Blaser et al. (1986). Seasonal variation in protein, ADF, NDF, and crude fat was evident in samples collected from the research pastures of the MARE Center (Wilson et al., 1997).

The overestimation of CHO-H by NSC (Figure 8.1b) may be due to increased soluble fiber content. Soluble fibers, including gums, mucilages and pectins, are found in the plant cell components (Van Soest et al., 1991) and are rapidly fermented in the hind gut of the horse.

Non-structural carbohydrate as determined "by difference" overestimated the CHO-H concentration of the starch and sugar supplement (CHO-H = 0, NSC = 17% DM, Figure 8.2a). Likewise, NSC may overestimate CHO-H for other cereal based concentrates. For pasture and hay samples (Figure 8.3), this difference was not as apparent. However, NSC overestimated CHO-H in samples from MARE Center pastures (Figure 8.1).

The sugar and starch supplement is dissimilar from both the fat and fiber supplement and the pasture, as indicated by Figure 8.5. Therefore, its inclusion in the overall equation is statistically aberrant. However, a wide variety of grain supplements traditionally used in horse diets were not sampled in this study. Future research may fill in the gap between the NSC/CHO-H content of the pasture, the fat and fiber supplement, the sugar and starch supplement, and validate or revise this equation.

Supplementation of rapidly growing pasture with traditional grain based concentrates may provide an excess of dietary hydrolyzable carbohydrate. When hydrolyzable carbohydrates are consumed in excess by horses, a portion may escape hydrolysis in the small intestine and be fermented in the large bowel. Rapid fermentation may increase lactic acid production and increase the risk of digestive disorders (Clarke et al., 1990). Thus, accurate and precise determination of non-structural carbohydrate, including distinction between hydrolyzable and fermentable fractions, is needed to complete the profile of feed and forage analysis for horses.

Implications

Traditional methods of non-structural carbohydrate determination "by difference" did not provide an adequate estimation of the hydrolyzable carbohydrate present, as estimated by direct analysis. Non-structural carbohydrate "by difference" may overestimate hydrolyzable carbohydrate by as much as 17% in cereal concentrates. Due to the nature of digestion in the horse, the opportunity for hydrolysis of carbohydrate prior to fermentation, precise measurement of the hydrolyzable fraction would be beneficial.

Literature Cited

Blaser, R. E., R. C. Hammes Jr., J. P. Fontenot, H. T. Bryant, C. E. Polan, D. D. Wolf, F. S. McClaugherty, R. G. Kline, and J. S. Moore. 1986. Forage-Animal Management Systems. Bulletin 86-7. Virginia Agricultural Experiment Station, Virginia Tech, Blacksburg, VA.

Blaxter, K. 1989. Energy Metabolism in Animals and Man. Cambridge University Press, New York.

Clarke, L. L., M. C. Roberts, and R. A. Argenzio. 1990. Feeding and digestive problems in horses. Vet. Clin. N. Amer. Equine Prac. 6:433–451.

Davis, R. E. 1976. A combined automated procedure for the determination of reducing sugars and nicotine alkaloids in tobacco products using a new reducing sugar method. Tobacco Science 20:139–144.

Gray, G. M. 1992. Starch digestion and absorption in nonruminants. J. Nutr. 122:172–177.

Hall, J. M. 1989. A review of total dietary fiber methodology. Cereal Foods World 34:526–528.

SAS. 1988. SAS/STAT® User's Guide. (Release 6.03). SAS Inst. Inc., Cary, NC.

SlideWrite. 1995. SlideWrite Plus for Windows. (Ver. 3.00). Advanced Graphics Software, Inc., Carlsbad, CA.

Smith, D. 1981. Removing and analyzing total nonstructural carbohydrates from plant tissue. University of Wisconsin-Madison Research Report R2107, Agricultural Bulletin Building, Madison.

Van Soest, P. J., J. B. Robertson, and B. A. Lewis. 1991. Methods for dietary fiber, neutral detergent fiber and nonstarch polysaccharides in relation to animal nutrition. J. Dairy Sci. 74:3583–3597.

Wilson, J. A., D. S. Kronfeld, W. L. Cooper, and D. Sklan. 1997. Seasonal variation in the nutrient composition of northern Virginia forages. p 336–331. Proc. 15th Equine Nutr. Physiol. Soc., Ft. Worth, TX.