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A model for assessing *Medicago Sativa* L. hay quality

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Abstract

A study was conducted to identify chemical parameters and/or models for assessing *Medicago sativa* L. (L) hay quality, using near infrared reflectance spectroscopy (NIRS) analysis and Cornell Net Carbohydrate and Protein System (CNCPS) milk prediction as a criterion of accuracy. Milk yield (MY) derived from the CNCPS model, by replacing the average L hay in a complete diet with 168 representative South African L hay samples, was used as a criterion to evaluate and/or develop models for L hay quality grading. The best single predictor of MY was the acid detergent fibre (ADF) content of L hay, which explained 67% of the measured variation. A multiple linear equation (Y = 64.18 – b1ADF – b2ash – b3lignin) explains 96% of the measured variation in MY. The relatively poor performance of crude protein (CP) ($r^2 = 0.04$) and other protein related parameters ($r^2 < 0.25$; adjusted-crude protein, ADF-CP, neutral detergent fibre-CP and soluble protein) in predicting MY suggests that protein content of L hay is an unreliable indicator of L hay quality. It is clear that MY derived from the CNCPS model by replacing L hay in a basal diet with others in the South African L hay population can be significantly predicted with high accuracy by the developed empirical model named lucerne milk value (LMV) consisting of only ADF, ash and lignin.

Keywords: Lucerne hay, CNCPS, lignin, ADF, ash, NIRS
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Introduction

Linn (1992) stated that forages vary in nutrient content more than concentrates and minerals. The dairy feed industry is the largest consumer of lucerne hay in South Africa. Consequently, its nutritive value and/or nutrient contribution for especially dairy cattle are important and need to be incorporated into lucerne hay quality assessment.

Since forages are predominantly used by ruminants as source of nutrition, forage quality is an expression of the characteristics that affect consumption, nutrient utilisation and resulting animal performance, therefore, production potential. Currently adopted models for assessing forage quality addresses only the chemical composition of the feed without considering interaction among other feeds (associated effects), rate of passage and physical characteristics. For this reason, forages are often referred to as functional feeds (Zinn & Ware, 2007). This stresses the necessity of considering the animal. One method available to evaluate the nutrition of dairy cows, and not so far exploited to assess the quality of forages, is the Cornell Net Carbohydrate and Protein System (CNCPS) (Tylutki *et al*., 2007).

The CNCPS is widely acknowledged as one of the most advanced ruminant feed evaluation models presently developed. The non-linear nature of the CNCPS model integrates nutrient intake, ruminal fermentation, intestinal digestion, absorption, and metabolism of chemical analyses and mathematical models with cattle requirements for each production situation (Fox *et al*., 2004). Fox *et al*. (2004) stated that the utility of the CNCPS is supported by the observation that various components have been adapted by the NRC (2001).

Several researchers assessed the ability of the CNCPS to predict lactation performance in dairy cows (Fox *et al*., 2004; Tylutki *et al*., 2007). In an evaluation with individual fed dairy cows, the CNCPS accounted for 90% of the variation in actual milk production with a 1.3% bias (Fox *et al*., 2004). The CNCPS is therefore a valuable aid to assess the quality of lucerne hay accurately according to animal performance. As previously mentioned, lucerne hay is mostly used for dairy nutrition and lucerne hay quality should accordingly be related to its milk production potential.

The usage of near infrared reflectance spectroscopy (NIRS) for the rapid and economical nutritional evaluation of lucerne hay is a very real need for the implementation of a practical lucerne hay evaluation model. Thus, the usefulness of applying NIRS in lucerne hay quality assessment will largely depend on the accurate prediction of the selected parameter's calibration equations, used in the quality model. The most robust NIRS calibration equations were identified and discussed by Scholtz *et al*. (2009) and should be considered when developing (modelling) and/or identifying an appropriate model for South African lucerne hay quality grading.

A study was conducted to identify chemical parameters and/or models for assessing lucerne hay quality, using NIRS analysis and CNCPS milk prediction as criterion of accuracy.

Materials and Methods

Six hundred lucerne (*Medicago sativa* L.) hay samples were collected from several commercial irrigation farms in the main lucerne producing areas in South Africa, which varied in location, soil characteristics (texture, organic matter, N content, pH) and farm management. Hundred and sixty eight samples that represented the South African lucerne hay population were selected with a NIR-Systems Model 5000 scanning monochromator (Foss NIR Systems, Silver Spring, MD, USA), analysed (chemical and *in vitro*) and near infrared reflectance spectroscopy (NIRS) calibration equations developed. The software for scanning, mathematical processing, calibration and statistical analysis with the NIR spectrophotometer was supplied by Infrasoft International® software 3.01 version (ISI, Port Matilda, PA, USA) (Scholtz *et al*., 2009).

A basal diet for lactating dairy cows was formulated, using a modified version of CNCPS (CNCPSv6) (AMTS.Cattle version 1.1.0.1, AMTS, LLC, 418 Davis RD, Cortland, NY, 13045, USA) (Tylutki *et al*., 2007). It was based on the milk production level, feed ingredients and dry matter intake (DMI) of a typical high producing South African commercial Holstein herd. The age of cows was assumed to be 42 months, body weight 700 kg, days pregnant 0, days since calving 60, body condition scoring (BCS) 3.00, average daily gain (ADG) 0.079 kg/d, milk production 45 L/d, milk fat 3.55%, and milk protein 2.88%. Ambient temperature was 20 °C and relative humidity (RH) was 50%, with no wind or other sources of environmental stress.

The basal diet consisted of hominy chop, ground maize, wheat bran, molasses, whole cotton seed, cotton oil-cake (OC), soya OC, sunflower OC, lucerne hay and a mineral-vitamin pre-mix. The lucerne hay used in the basal diet was selected from the average chemical values of 168 selected lucerne hay samples.

The basal diet formulation was based on the following criterion namely: metabolisable protein (MP) from bacteria >50% of total MP supply; non fibre carbohydrate (NFC) <40%; physical effective NDF (peNDF) and predicted ruminal pH above the minimum requirement of 22% and 6.4, respectively; rumen ammonia- and peptide-balance >100%, respectively; milk urea nitrogen (MUN) between the allowable 12 – 18 mg/dL; methionine (Met) and lysine (Lys) retained >2.2% and >6.8% of MP supply, respectively whereas the Lys : Met ratio was maintained around 3 : 1; metabolisable energy (ME) and MP allowable milk within 2 : l of each other.

The lucerne in the basal diet was replaced with the rest of the South African lucerne hay population described above. A diet with lucerne hay as sole roughage source was formulated to evaluate the effect of lucerne hay quality on ME and MP allowable milk. Hence, a total of 168 simulations were run to obtain milk yield (MY) values. A DMI of 25 kg/d were maintained during the simulation to cancel the effect of DMI. The effect of replacing lucerne hay in the basal diet on the protein fractions and MUN were addressed by using the lowest ME or MP allowable milk yield (kg/d) for each lucerne hay sample.

Replacing lucerne hay in the basal diet with the rest of the South African population also resulted in a variation in nutritional factors like peNDF, NFC and pH that could influence milk composition (milk fat) and the occurrence of sub-acute ruminal acidosis (SARA). Rectifying changes in these nutritional factors when replacing lucerne hay in the basal diet seemed to be unnecessary, as the maximum NFC values and minimum pH values were acceptable and most of the lucerne hay used resulted in diets that generally satisfied the peNDF needs of dairy cows (Mertens, 1997; Fox *et al*., 2004; Tylutki *et al*., 2007). Furthermore, the buffering capacity of lucerne hay and its high pectin content (Calberry *et al*., 2003) would induce a smaller effect of peNDF on ruminal pH. Considering changes in milk fat when evaluating the quality of lucerne hay is also not so important as many milk buyers in South Africa have no prerequisite regarding milk fat content. Therefore rectifying changes in these nutritional factors by adding feeds in the diet were unnecessary and would result in confounded effects.

Statistical analyses were performed using SAS 9.1.3 Service Pack 4 (2002-2003). Simple and multiple regression analyses were done separately for individual sources of variation. In the multiple regression analyses, forward selection was used in the stepwise regression calculations. This method starts with no variables in the model and adds variables. Variables were only retained in the model when they significantly ($P < 0.05$) contributed to the accountable variances. Expressions of the proportion of the sum of squares for the dependent variable explained by these regressions were calculated as r^2.

Results and Discussion

From Table 1 it seems that the variation in lucerne hay quality according to ME and/or MP milk yield (MY) range from 35.7 to 48.7 kg/d. Thus, normal variance of MP and ME in South African lucerne hay alone can vary milk production of high producing Holstein cows by 13.0 kg per cow per day (Table 1).

Table 1 Nutritional indicators and milk response of 168 SA lucerne hay based diets

	Minimum	Mean	Median	Maximum	Range	SD[1]	CV[2]
Milk yield [3] (kg/d)	35.72	43.01	43.58	48.69	12.97	2.78	6.46
Metabolisable energy milk (kg/d)	35.72	43.01	43.58	48.69	12.97	2.78	6.46
Metabolisable protein milk (kg/d)	38.42	45.74	46.02	52.76	14.34	2.88	6.30

[1] Standard deviation
[2] Coefficient of variation
[3] First limiting (metabolisable energy or metabolisable protein) allowable milk production

The best single predictor of MY was the acid detergent fibre (ADF) content of lucerne hay, which explained 67% of the measured MY variation (Table 2). An improvement ($r = 0.96$) has, however, been accomplished by combining ADF, ash and lignin in a multiple regression and/or model named lucerne milk value (LMV) ($Y = 64.18 – b1ADF – b2ash – b3lignin$) (Table 2). According to the standards of Williams (2001) this model includes accurate and acceptable NIRS calibration equations ($r^2 > 0.92$; ratio of prediction to deviation >3.0; standard error of cross validation <1.10) for lucerne hay quality in practice.

Table 2 Regression equations for predicting milk yield (MY) and the coefficient of determination (r^2) between dependent and independent variables in lucerne hay

Independent variate (X)	Dependent variate (Y)	r^2	Regression equation $Y = b_0 + b_1X_1 + b_2X_2 + b_3X_3 + b_4X_4$
Chemical and digestibility			
Acid detergent fibre (X_1)	Milk Yield	0.67	$Y = 60.72 - 0.54X_1$
Ash (X_2)		0.87	$Y = 64.40 - b1X_1 - b2X_2$
Lignin (X_3)		0.96	$Y = 64.18 – b1X_1 – b2X_2 - 0.90X_3$
NDFD24 [1] (X_4)		0.97	$Y = 64.16 – b1X_1 – b2X_2 - 0.85X_3 - 0.09X_4$

[1] *In vitro* neutral detergent fibre digestibility at 24 hours.

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[1] *In vitro* neutral detergent fibre digestibility at 24 hours.

According to Allen & Mertens (1988) NDFD is a function of the potentially digestible fraction, its rate of digestion and rate of passage. It was therefore expected to correlate highly with MY. In contrast with these expectations, NDFD24 and NDFD48 explained only 21 and 10%, respectively, of the variance observed in MY. Furthermore, the accountable variance of 96%, based on pure chemical parameters (ADF, ash and lignin), could only be marginally improved to 97% by the inclusion of a digestibility parameter (NDFD24) (Table 2).

Even though ADF captures less of the structural fibre, it (ADF) out-performed neutral detergent fibre (NDF) ($r^2 = 0.61$) in estimating available energy for milk production (MY). Accordingly, the regression equation based on ADF explained more of the variance in MY than *in vitro* digestibility parameters ($r^2 < 0.52$). Therefore, it seems from the results of the present study that chemical extractions could be more closely related to animal performance than *in vitro* estimates of digestion.

Much emphasis has been placed on CP content of lucerne hay in the past as the ultimate predictor of quality. The relatively poor performance of CP ($r^2 = 0.04$) and other protein related parameters ($r^2 < 0.25$; ACP, ADF-CP, NDF-CP and SP) in predicting MY suggested that the protein content of lucerne hay, as such, is an unreliable indicator of production potential, therefore of lucerne hay quality.

Conclusions

The evaluation of different parameters for assessing lucerne hay quality revealed large differences in the accuracy of prediction as measured by MY. From the results of the present study it seems that the ADF of lucerne hay in a dairy diet is the single pure chemical parameter most closely related to milk yield production potential of dairy cattle. A marked improvement in the accuracy of milk yield prediction and therefore lucerne hay quality occur by including ADF, ash and lignin in a multiple linear equation.

The relative poor performance of the protein parameters (ACP, ADF-CP,NDF-CP and CP) in predicting MY suggests that protein content of lucerne hay, in general, is an unreliable indicator of production potential (MY) and/or lucerne hay quality.

It is clear that milk yield derived from the CNCPS model by replacing lucerne hay in a basal diet with others in the South African lucerne hay population can be significantly predicted with high accuracy by the developed empirical model (LMV) consisting of only ADF, ash and lignin. Furthermore, accurate NIRS calibration equations for these parameters (ADF, ash and lignin) has proven to successfully assist in producing an accurate, rapid and economical estimate of MY potential for lucerne hay. These calibration equations and LMV could be used to apply an accurate grading system for lucerne hay in practice.

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