

The use of indirect methods for the prediction of lucerne quality in the first cut under the conditions of Central Europe

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ABSTRACT: The goal of this study was to compare the growing degree-days (GDD) and predictive equations for alfalfa quality (PEAQ) for the prediction of lucerne quality and to test their accuracy and suitability in the first cut period in Central Bohemia. Several additional stand parameters were verified in order to increase the accuracy of the quality estimate under these environmental conditions. In 2004–2007, the measurement and sampling were repeatedly realized from the late vegetative to the early bloom stage in six replications. For the GDD model across the years, the obtained R^2 for NDF, ADF and CP were 0.40, 0.57 and 0.65, respectively. It seems that the forage quality response to accumulated GDD was related to the stand development and it could be the reason for low R^2 across all the years. For the PEAQ model, R^2 were 0.62, 0.92, and 0.85, respectively. Similarly like in the GDD model, the effect of stand development across the years on changes in the slopes of equations was observed. The accuracy of the model combination was not higher in comparison with the PEAQ model. The count of stems per plant, density of stems per m^2 , dry matter yield and average stem weight improved the NDF content prediction within a four-year period. In these models, variables which represent the stand development should be taken into account.

Keywords: forage; alfalfa; quality; stand; fibre

The timing of spring forage harvest is critical for obtaining optimal quality for animal production. For forage that serves as the primary fibre source in the diet, NDF is the principal forage quality variable of concern (Parsons et al., 2006a). Some predictive equations can be used to estimate the forage quality of lucerne, assisting the producers in decision making at harvest time. Parsons et al. (2006b) described an ideal method for estimating lucerne quality in the field as a harvest decision aid must be quick, simple, inexpensive, and consistent across all harvests during the season and across a wide range of environments.

A number of methods have been developed to estimate lucerne NDF, including models based on weather, chronological age and plant morphology (Fick et al., 1994). These parameters are related not only to NDF or ADF but also to CP, DOM, and IVDDM (Sulc et al., 1997; Rinne and Nykanen, 2000; Mitchell et al., 2001). Near infrared reflectance spectroscopy (NIRS) belongs to indirect methods (Mika et al., 2003) but this method is not so suitable for field conditions, moreover, special instrumentation is needed.

The most widely used of these methods are predictive equations for alfalfa quality (PEAQ). This

method is based on the length of the tallest stem and the stage of the most mature stem in the sample (Hintz and Albrecht, 1991). The equations also performed well across the spring and summer growth cycles (Sulc et al., 1997) although lucerne quality varied among growth cycles during the year (Niwinska et al., 2005), which is connected with changes in the leaf stem ratio (Tyrolová and Výborná, 2008). Similarly Skládanka et al. (2008) documented changes in fibre content in forage of grasses at the end of the growing season. These equations have been developed for many regions of the USA. Results indicated some bias when the equations were used outside the state of development; however, the prediction errors were sufficiently low to suggest the PEAQ equations are robust over a wide range of environments (Parsons et al., 2006a). Parsons et al. (2006b) described some modified equations, based only on lucerne height.

Growing degree-days (GDD) are a temperature-derived index representing the amount of heat to which plants are exposed. This method has been used successfully in maize development, but has had a mixed success in the perennial types of forage (Sulc et al., 1999). The GDD for lucerne are calculated as follows. Average the maximum and minimum temperature for each day (24 h period) beginning March 1, subtract the base temperature ($41^{\circ}\text{F} = 5^{\circ}\text{C}$) and sum up the growing degrees for all days that have a positive number (Allen and Beck, 1996). The GDD are not reliable when the soil moisture is inadequate, which often occurs during the second cutting growth. It is so presumably because

forage growth is limited more by soil moisture than by heat units. Neither GDD nor PEAQ adequately predicted NDF for the third cutting lucerne (Lee et al., 2002). Similarly, the chronological age of forage relates to quality in spring growth, but it is inconsistently related to quality in regrowth (Sulc et al., 1999). In Europe, the GDD method has proved to be an accurate predictor of grass digestibility under Finnish conditions (Rinne and Nykanen, 2000). The base temperature is 5°C as published by Allen and Beck (1996).

The above discussed methods are quick, easy and their accuracy is acceptable under the USA conditions (Owens et al., 1995; Allen and Beck, 1996; Sulc et al., 1997). Since no single method will ever result in a perfect prediction of lucerne quality in the future, a combination of methods may be most acceptable (Sulc et al., 1999). Allen and Beck (1996) suggested that GDD in combination with the plant height and the maturity stage might be more accurate for estimating lucerne NDF than using GDD alone. Some additional variables for improving accuracy in models were used by Parsons et al. (2006a) in a lucerne grass mixture. As noted by Hakl et al. (2006), the stand structure could influence the fibre content of lucerne but this effect is low in dense stand. It is important to understand that any method used to predict or estimate forage quality will have an error in relation to wet chemistry analyses (Sulc et al., 1999).

In the Czech Republic, these methods have not yet been tested for any perennial forage crops. The goal of this study was to assess preliminary equations of these methods and to test their accuracy and suitability for

Table 1. Descriptions and ranges of variables evaluated as potential predictors of lucerne quality in lucerne stand

Variables	Description	Min.	Max.
ASW	average stem weight (g)	2	21
D	stand density (plants/m ²)	75	700
DMY	dry matter yield (kg/ha)	2 000	17 775
GDD	accumulated growing degree days ($^{\circ}\text{C}$), base 5°C	193	578
MSL	height of the tallest lucerne stems in the sample area (cm)	29.2	143.0
S	morphological stage of development of the most mature lucerne stems in the sample area	2	5
SPP	count of stems per plant (pcs/plant)	2.2	15.3
TSC	total stem count (pcs/m ²)	375	2 375
Y	year of vegetation	2	5

Table 2. Lucerne developmental stage used to assign a numerical value to the most mature stems in the sampling area, codes of stages and descriptions are according to Kalu and Fick (1981)

Developmental stages	Code	Description
Late vegetative	2	stems length > 300 mm, no visible buds
Early bud	3	1 to 2 nodes with visible buds
Late bud	4	> 2 nodes with visible buds
Early bloom	5	1 node with at least 1 open flower

lucerne in the first cut period in Central Bohemia. In these models, we also verified some additional stand parameters for increasing the accuracy of the quality estimate in our environmental conditions.

MATERIAL AND METHODS

In spring 2003, the lucerne experiment was established at the Czech University of Life Sciences in Prague in Central Bohemia (286 m above sea level, 50°08' N, 14°24' E). The long-term annual temperature is 9.3°C and precipitation 510 mm.

The experimental plants (*Medicago sativa* L.) consisted of two lucerne cultivars (ŽE XLII, ŽE XLV) and the Jarka variety grown in two replications on completely randomised blocks. In 2004–2007, the measurements and samplings were repeatedly realized from the late vegetative to the early bloom stage in the first cut per each treatment in a 20 × 20 cm square. Stand density, total stem count, dry matter yield, and count of stems per plant were assessed for each sample. The height of the tallest lucerne stem in the sample area was measured to the terminal point of the stem. Abbreviations of the variables, their descriptions and ranges are summa-

Table 3. Means of yield and stand structure parameters

Year	S	D (plants/m ²)		TSC (pcs/m ²)		MSL (cm)		SPP (pcs/plant)		ASW (g)	
		mean	SE	mean	SE	mean	SE	mean	SE	mean	SE
2004	2	412	56	1 254	166	33.0	1.5	3.07	0.17	3.05	0.42
	3	433	68	1 162	170	48.9	2.3	2.73	0.10	3.59	0.37
	4	454	50	1 437	146	63.0	4.2	3.21	0.26	5.55	0.86
	5	354	50	1 037	90	73.8	4.1	3.05	0.24	7.48	0.28
2005	2	337	37	1 537	150	45.3	1.7	4.60	0.24	3.69	0.46
	3	204	16	979	93	60.3	2.3	4.81	0.31	5.89	0.53
	4	279	18	970	135	74.1	3.0	3.47	0.45	8.68	0.86
	5	258	11	941	58	106.1	3.2	3.70	0.32	12.33	1.13
2006	2	154	12	1 033	295	60.8	1.8	6.62	1.64	6.24	1.28
	3	187	11	1 341	106	81.1	1.4	7.19	0.50	7.62	0.78
	4	192	18	1 088	66	98.8	5.5	5.85	0.48	11.62	0.94
	5	150	18	800	100	120.3	6.1	5.34	0.27	14.83	1.70
2007	2	117	14	933	129	52.8	2.1	8.23	1.12	6.02	0.51
	3	125	17	1 133	44	70.8	3.4	9.96	1.45	8.73	0.99
	4	121	10	913	70	79.3	1.8	7.81	0.94	10.44	1.05
	5	117	17	838	91	94.8	2.6	7.60	0.87	14.42	1.65

SE = standard error; $n = 6$; for the description of variables see Table 1

Table 4. Means of forage yield and quality

Year	S	Date	GDD (°C)	DMY (kg/ha)		CP (%)		ADF (%)		NDF (%)	
				mean	SE	mean	SE	mean	SE	mean	SE
2004	2	12.5.	324	3 516	207	24.67	0.39	25.41	0.99	30.21	0.66
	3	21.5.	405	3 879	417	22.58	0.53	29.69	0.77	34.46	0.96
	4	31.5.	471	7 550	729	18.94	0.54	33.66	1.06	37.12	1.32
	5	9.6.	578	7 770	764	20.28	0.71	34.86	1.30	40.36	1.58
2005	2	4.5.	262	5 512	744	24.23	0.99	26.07	4.59	40.72	2.43
	3	19.5.	341	5 608	410	23.50	0.40	30.00	1.05	45.38	1.02
	4	25.5.	389	8 108	1 052	21.40	0.61	32.93	0.66	43.10	1.81
	5	7.6.	567	11 512	1 184	16.57	0.81	43.87	1.15	52.87	1.25
2006	2	10.5.	265	5 400	1 106	26.11	0.84	29.82	1.28	34.72	0.76
	3	16.5.	324	10 279	1 415	23.78	0.47	32.70	1.07	36.85	0.76
	4	25.5.	416	12 616	1 269	20.63	0.43	36.92	0.52	43.17	0.51
	5	9.6.	512	11 895	1 817	16.47	0.80	41.62	2.01	47.68	1.80
2007	2	24.4.	193	5 345	544	25.12	0.81	26.13	0.95	30.08	1.28
	3	2.5.	258	9 775	951	21.55	0.42	31.77	0.86	35.67	0.68
	4	7.5.	300	9 663	1 433	21.23	0.57	35.28	1.77	40.12	2.41
	5	16.5.	394	11 446	684	19.16	0.55	35.32	1.07	43.77	1.68

SE = standard error; $n = 6$; for the description of variables see Table 1

rized in Table 1. The lucerne maturity categories of Kalu and Fick (1981) (Table 2) were used to assign a numerical value to the most mature stem in the sample area. All samples were oven-dried at 60°C, homogenised to a particle size of 1 mm, and analysed for crude protein (CP), neutral detergent fibre (NDF), and acid detergent fibre (ADF) content. CP was quantified using the Kjeldahl procedure for N determination (% CP = % N × 6.25; AOAC, 1990). The NDF and ADF contents were determined according to Van Soest et al. (1991). GDD for each harvest date were calculated according to Allen and Beck (1996). Direct and stepwise linear regression analyses were performed using Statistica 6.1 (StatSoft, 2003).

RESULTS AND DISCUSSION

The means and standard errors of measured parameters are shown in Tables 3 and 4. You can see increasing DMY and MSL during the follow up first cut periods. Within a four-year period, increas-

ing SPP was caused by a natural decrease of stand density and subsequent plant development which is associated with changes in the root traits (Hakl et al., 2007). The changes in lucerne quality during the first cut periods correspond with changes described by Homolka et al. (2008). No significant differences among lucerne entries in forage quality were detected, therefore this factor was not included in subsequent analyses.

As noted by Lee et al. (2002), the GDD is not reliable when the soil moisture is inadequate. During the first cut period, this site did not show any signs of moisture deficit in all experimental years. The models of the GDD method are shown in Table 5. The R^2 and standard error of prediction (SEP) values varied from one year to the next. This is in accordance with Allen and Beck (1996), who reported that prediction equations developed in one year were not stable across the years. Cherney (1995) found that GDD prediction equations of NDF content developed for different data sets had considerably different intercepts, but the slopes of the equations were relatively consistent. In our ex-

Table 5. Relating lucerne quality (y ; NDF, ADF, CP; %) and accumulated growing degree-days (x ; GDD; °C) in the first cut period (linear regression, $y = a + bx$)

Y quality	Year	X = GDD model	SEP	R^2	P -value	n
NDF	2004	18.06 + 0.039 GDD	2.81	0.65	0.0000	24
	2005	30.59 + 0.038 GDD	4.33	0.52	0.0000	24
	2006	19.91 + 0.055 GDD	2.58	0.81	0.0000	24
	2007	17.85 + 0.068 GDD	4.01	0.63	0.0000	24
	2004–2007	26.77 + 0.035 GDD	4.96	0.40	0.0083	16*
ADF	2004	14.06 + 0.038 GDD	2.72	0.65	0.0000	24
	2005	10.16 + 0.059 GDD	5.73	0.59	0.0000	24
	2006	17.23 + 0.048 GDD	3.11	0.69	0.0000	24
	2007	19.27 + 0.045 GDD	3.42	0.50	0.0001	24
	2004–2007	19.90 + 0.035 GDD	3.54	0.57	0.0008	16*
CP	2004	30.02 – 0.019 GDD	1.88	0.49	0.0001	24
	2005	31.79 – 0.026 GDD	1.79	0.75	0.0000	24
	2006	36.37 – 0.039 GDD	1.55	0.85	0.0000	24
	2007	29.77 – 0.028 GDD	1.57	0.65	0.0000	24
	2004–2007	29.40 – 0.021 GDD	1.77	0.65	0.0001	16*

*for elimination of heterogeneity on sampling days the average values from Table 3 and 4 were used

SEP = standard error of prediction, intercepts (a) and slopes (b) significant at $P < 0.05$

Table 6. Relating lucerne quality (y ; NDF, ADF, CP; %) and stage of development (x_1 ; S; code 2–5) and maximal stem length (x_2 , MSL; cm) in the first cut period (PEAQ method, linear regression, $y = a + b_1x_1 + b_2x_2$)

Y quality	Year	X model	SEP	R^2	P -value	n
NDF	2004	21.95 – 0.07 S + 0.30 MSL	1.56	0.90	0.0000	24
	2005	32.95 – 0.99 S + 0.22 MSL	4.54	0.49	0.0008	24
	2006	23.83 + 3.67 S + 0.04 MSL	2.72	0.80	0.0000	24
	2007	26.58 + 7.06 S – 0.19 MSL	3.77	0.69	0.0000	24
	2004–2007	24.11 + 1.85 S + 0.13 MSL	4.11	0.62	0.0019	16*
ADF	2004	17.99 + 0.05 S + 0.23 MSL	1.92	0.83	0.0000	24
	2005	12.86 + 1.05 S + 0.23 MSL	5.95	0.58	0.0001	24
	2006	22.14 + 4.64 S – 0.03 MSL	3.20	0.69	0.0000	24
	2007	25.30 + 5.10 S – 0.15 MSL	3.14	0.60	0.0000	24
	2004–2007	16.89 + 1.67 S + 0.14 MSL	1.61	0.92	0.0000	16*
CP	2004	27.75 – 1.17 S – 0.04 MSL	1.77	0.57	0.0000	24
	2005	30.54 – 0.63 S – 0.10 MSL	1.92	0.72	0.0000	24
	2006	33.30 – 2.90 S – 0.02 MSL	1.65	0.84	0.0000	24
	2007	27.20 – 2.28 S + 0.03 MSL	1.60	0.65	0.0000	24
	2004–2007	29.99 – 2.01 S – 0.02 MSL	1.20	0.85	0.0000	16*

*for elimination of heterogeneity on sampling days the average values from Table 3 and 4 were used

SEP = standard error of prediction, intercepts (a) and slopes (b_1 , b_2) significant at $P < 0.05$

periment, the intercepts of equations for the NDF prediction between years were relatively consistent, except for the year 2005. The slopes increased from 0.039 in 2004 to 0.068 in 2007. In 2004, the lowest slopes were also recorded for ADF and CP. We can assume that the utilization of heat units for fibre production varied in dependence on the stand structure when an older stand with more developed plants increased the fibre content faster than a younger stand with less developed plants. This effect was most obvious in the case of NDF with continually increasing slopes from 2004 to 2007. It is possible to conclude that the forage quality response to accumulated GDD was related to stand development in the four-year period. It could be the reason for low R^2 and high SEP values of prediction equations across all the years. In this case, the best estimate was recorded for CP where the R^2 value was 0.65.

The models of PEAQ are shown in Table 6. We used the 4-stage scale according to Table 2. In contrast to GDD models, the slopes in equations of each year were not always significantly different from zero. Similarly like in the GDD model, the R^2 and SEP values varied from one year to the next. The prediction equations across all the years were more accurate than the GDD models. The original equations according to Hintz and Albrecht (1991) reached the R^2 values of 0.89 for NDF ($= 16.89 + 0.81 S + 0.27 \text{ MSL}$), 0.88 for ADF ($= 11.57 + 0.79 S + 0.21 \text{ MSL}$) and 0.74 for CP ($= 30.71 - 0.89 S - 0.09 \text{ MSL}$). Sulc et al. (1997)

evaluated the performance of PEAQ across five states and presented the R^2 value 0.74 for NDF ($= 23.67 + 0.41 S + 0.21 \text{ MSL}$) and 0.73 for ADF prediction. According to Owens et al. (1995), the CP was not predicted accurately using PEAQ. In the first cut, they obtained the R^2 values of 0.83 for NDF ($= 18.19 + 1.44 S + 0.21 \text{ MSL}$), 0.78 for ADF ($= 11.92 + 1.11 S + 0.18 \text{ MSL}$) and 0.37 for CP ($= 29.63 - 0.87 S - 0.09 \text{ MSL}$). In our experiment, the R^2 values were 0.62, 0.92, and 0.85, respectively. Similarly like in the GDD model, the effect of stand development across years on changes in slopes of the equations was observed. This effect was most obvious in the case of fibre with increasing slopes of maturity and decreasing slopes of MSL from 2004 to 2007 with opposite trends for CP. Generally, it seems that across years the slopes for stage were higher and for MSL they were lower in comparison with the equations mentioned above.

The combinations of models are shown in Table 7. The combination of GDD and PEAQ models was better for all quality parameters than GDD alone, which is in accordance with suggestions of Sulc et al. (1999) that the combination of methods might be most acceptable. The accuracy of the model combination was not higher in comparison with the PEAQ model. The highest accuracy was observed for ADF, the lowest for NDF. The GDD model alone is less accurate than PEAQ but, according to Sulc et al. (1999), it eliminates the need to sample a field, and therefore eliminates the potential for lucerne sampling error.

Table 7. Combination of models and stepwise regression for the prediction of lucerne quality (NDF, ADF, CP; %); linear regression

	Model*	SEP	R^2	P-value
Combination of GDD and PEAQ				
NDF	$22.46 + 0.15 \text{ MSL} + 0.259 S + 0.015 \text{ GDD}$	4.16	0.64	0.0054
ADF	$15.28 + 0.16 \text{ MSL} + 0.117 S + 0.014 \text{ GDD}$	1.37	0.94	0.0000
CP	$30.61 - 0.03 \text{ MSL} - 1.41 S - 0.006 \text{ GDD}$	1.19	0.86	0.0000
Stepwise regression				
NDF	$48.01 + 0.017 \text{ TSC} - 3.72 \text{ SPP} - 0.08 \text{ DMY} + 2.95 S + 2.87 \text{ ASW}$	3.20	0.82	0.0017
ADF	$15.24 + 0.16 \text{ MSL} + 0.015 \text{ GDD}$	1.32	0.94	0.0000
CP	$29.72 - 2.31 S$	1.18	0.84	0.0000

*for the description of model variables see Table 1

SEP = standard error of prediction, intercepts and slopes significant at $P < 0.05$, $n = 16$

From other variables, the stepwise algorithm included SPP, DMY, TSC and ASW to models for NDF prediction whilst ADF was the best for the model using MSL and GDD. CP prediction was the best using only the stage of development (Table 7). The accuracy of stepwise models was similar to the PEAQ model except for NDF where the R^2 value increased to 0.82. The significant slopes of selected variables indicate their importance for fibre prediction. The SPP variable is a function of D and TSC. It corresponds with Šantrůček (1989), who stated that stand density can influence the fibre content although this effect is low in a dense stand (Hakl et al., 2006). In our experiment, these variables represent a level of stand development where they correspond with the changes of stand in a four-year period. It seems that the accuracy of the models could be influenced by these changes. The inclusion of variables which characterized the stand development in the models can improve the accuracy of the fibre estimate and thus it should be taken into account.

CONCLUSIONS

The PEAQ method seems to be a promising method for the lucerne quality prediction under the conditions in Bohemia. The GDD method was less accurate across the years and the combination with the PEAQ did not improve the quality of the models. However, the GDD method eliminates the need to sample a field, so it would be suitable as the signal method for starting with field evaluation. In both methods, the accuracy of estimates was the lowest for NDF prediction. According to our opinion, the combination of developmental stage and stand height or GDD seems to be a promising method for predicting harvest dates in the Czech Republic and thus suitable for field experiments as well as for farm use. To develop a functional model, experiments on many sites are needed with a follow up validation of this model. In our opinion, the variables which represent stand development should be taken into account in these models mainly for NDF prediction.

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Received: 2009–04–08

Accepted after corrections: 2009–12–14

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- **Books and article within edited books, proceedings**

Spally M.R., Morgan S.S. (1989): *Methods of food analysis*. 2nd Ed. Elsevier, New York, 682 pp.

Kalab J. (1995): Changes in milk production during the sexual cycle. Hekel K. (ed.): *Lactation in Cattle*. Academic Press, London, UK, 876–888.

Janson L., Ahlin K.A. (1992): Postpartum reproductive performance in cattle selected for high and low fat content. In: *Proc. 43rd Annu. Mtg. European Association for Animal Production (EAAP)*. Madrid, Spain, 93–95.

- **Patent:**

Harred J.F., Knight A.R., McIntyre J.S., inventors; Dow Chemical Co., assignee. 1972 Apr 4. Epoxidation process. U.S. patent 3, 654, 317.

- **Dissertation:**

Smith D.E. (1988): Lipid oxidation at very low water activities. PhD Diss. Ithaca, NY: Cornell University. Available from: University Microfilms, Ann Arbor, MI: ABD62-83. 210 p.

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Revised February 2010