Colour measurement of hay using near infrared spectroscopy

P.C. Flinn

Agriculture Victoria, Pastoral and Veterinary Institute, Private Bag 105, Hamilton, Vic. 3300, Australia.

A.B. Blakeney

NSW Agriculture, Yanco Agricultural Institute, Yanco, NSW 2703, Australia.

Introduction

The gross value of the Australian hay industry is worth around A$700 million per year. Victoria, occupying 3% of the land mass, accounts for 40% of all hay produced in Australia and 55% of all pasture hay. There is an increasing trend towards trading hay on the basis of objective criteria such as metabolisable energy and protein content, and near infrared (NIR) spectroscopy is playing an important role in this. However, the industry still places importance on subjective criteria such as colour, aroma and texture. No quantitative measure of colour is currently used for hay in Australia, and this study was aimed at developing an NIR calibration for hay colour, based on measurements with a colour instrument.

Materials and methods

The study utilised 404 hay samples, representing a wide range of species, quality and environments across southern Australia, analysed by the FEEDTEST service during the 1994/95 hay season. All samples were oven-dried at 60°C and ground to pass a 1 mm screen in a cyclone mill. NIR spectra were collected on all samples as log 1/*R*, from 400 to 2500 nm, using a model 6500 scanning monochromator (NIRSystems Inc., Silver Spring, MD). A representative subset of 192 samples was chosen using the CENTER and SELECT algorithms contained in ISI software (Infrasoft International, Port Matilda, PA).

Colour was measured on the 192 samples in L*a*b* (CIE 1976) colour space[1] using the 10° observer and DL65 on a Micromatch 2000 diode array spectrometer fitted with an integrating sphere and d/8° viewing geometry. L* indicates lightness, and a* and b* are the chromaticity co-ordinates, where a* is the red–green axis (+a* is the red direction, –a* is the green direction), and b* is the yellow–blue axis (+b* is the yellow direction, –b* is the blue direction). NIR calibrations were derived for L*, a*, b* and reflectance at 520 nm (maximum greenness) using modified partial least squares (PLS) and first derivative math treatment (1,4,4,1). Cross-validation was carried out by splitting the data into four segments.

Results and discussion

Figure 1 shows the NIR spectra of two hay samples with widely differing lightness. The green clover hay had a lower L* than the pale yellow oaten hay and exhibited a stronger absorption

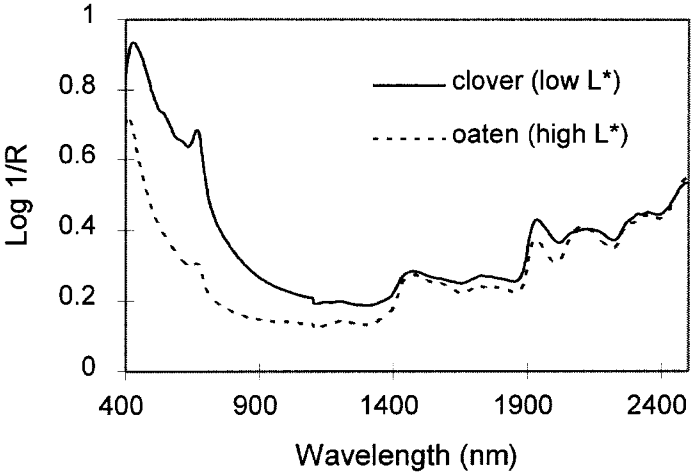

Figure 1. NIR log 1/*R* spectra of two hay samples with differing lightness.

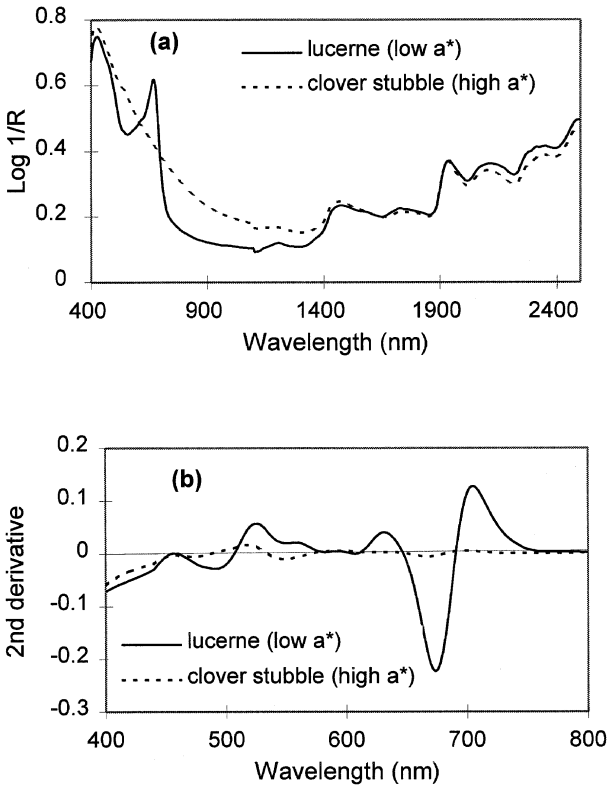

Figure 2. NIR spectra of two hay samples with widely differing red–greenness: (a) log 1/*R* spectra and (b) second derivative log 1/*R* spectra.

Table 1. Mean, range and standard deviation in colour measurements of a diverse population of hay samples, together with NIR calibration statistics.

Variable	*N*	Mean	Range	*SD*	*SECV*	R^2	*SECV/SD*
L*	186	60.8	49.5–71.1	4.36	1.02	0.94	0.23
a*	179	0.3	–5.5–5.2	2.23	0.28	0.98	0.13
b*	185	22.5	18.0–28.3	2.42	0.59	0.94	0.24
R(520)	186	27.1	13.1–39.0	4.88	1.16	0.94	0.24

N = number of samples; *SD* = standard deviation of values across population; *SECV* = standard error of cross-validation; R^2 = coefficient of determination; L* = lightness (0 black, 100 white); a* = red–greenness (+ red, –green); b* = yellow–blueness (+ yellow, –blue; *R*(520) = reflectance at 520 nm (colour instrument).

(higher log 1/*R*) in the "red" region (674 nm), thus appearing darker and greener to the human eye.

Figures 2 (a) and 2(b) show the NIR spectra (log 1/*R* and second derivative respectively) of two hay samples with widely differing red–greenness. The a* values for the green lucerne hay and the brown clover stubble were –4.55 and 5.18 respectively. This resulted in the lucerne sample absorbing strongly in the "red" region (674 nm).

The NIR calibration statistics for the colour measurements made are shown in Table 1.

High calibration accuracy (high R^2, low *SECV*, *SECV/SD* ≤0.3) was obtained for all four colour measurements. When the visible region was excluded and spectral data from only the "NIR region" (1100–2500 nm) was used, calibration accuracy was lower (*SECV* and R^2 were 1.80 and 0.84, 1.33 and 0.65, 1.18 and 0.78 and 2.09 and 0.82, respectively). This could be expected, but there was still some correlation with the colour measurements.

In practice, the hay industry needs a simple "greenness index", similar in principle to the flour colour index used in the flour milling industry.[2] However, for adequate description of hay colour, the index should include all three colour parameters (L*, a* and b*) because of the three-dimensional nature of colour. This would be superior to using a single reflectance measurement at, for example, 520 nm (which was the point of maximum greenness as measured on the colour instrument).

Shenk[3] reported an *SECV* of 0.2 and an R^2 of 0.98 for a "green/brown" colour index of US hay. Efforts are currently underway to reach agreement between the USA and Australia on a common index to describe hay colour. Although colour is not a reliable indicator of the feeding value of hay, its prediction by NIR, along with other more important criteria, is a useful addition to a rapid analytical package and can be used as an educational tool for the hay industry as an alternative to visual assessment.

References

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3. J.S. Shenk, *Proc. XVII Intl. Grassld. Congr.*, pp. 591–592 (1993).