USE OF REFLECTANCE SPECTROSCOPY TO PREDICT ULTIMATE PH OF BEEF MEAT

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Keywords: pH, visible spectroscopy, multivariate analysis

Introduction

Much research has investigated the relationship between muscle pH and temperature with some physical characteristics (Bendall, 1979; Devine and Graafhuis, 1995), since pH measurement is an important tool for optimising meat quality (Tompson, 2002). pH is normally measured using a glass electrode, but fat and protein material tends to contaminate it during use compromising the accuracy of the data, so recently some authors have been studying alternative methods to measure pH.

In particular, ultimate pH seems to affect the ageing rate of meat and of cytoskeleton protein degradation. This also produces a change in the capacity of meat to absorb and reflect light, causing modifications in the reflectance spectrum, so the analysis of visible spectrum alternative methods could give some information on ultimate pH in meat, which is a complex matrix (Swatland, 1989). The aim of this paper is to evaluate the possibility of estimating the ultimate pH and the rate of pH fall using the visible spectrum of reflectance.

Materials and Methods

The experiment was carried out on 62 entire males from 3 different Italian cattle breeds: 20 Chianini (CN), 21 Marchigiani (MG) and 21 Piemontesi (PD). At slaughter, carcasses were chilled at 4° C for 24 hours, then the *Longissimus thoracis* muscle (between 7th and 10th rib) was removed from the left side of the carcass and stored in a vacuum package at 2°C±1°C. At 24, 48, 72, 120, 192 hours the sample was extracted from the package and, after removing the external slice, cleaned by water and exposed to oxygen for 1 hour. pH values (obtained as the mean of 4 determinations) with electrode penetration through meat using a pH meter (Hanna Hi 98240) and visual reflectance spectra between 360-740 nm (in steps of 10nm), using a reflectance spectrophotometer (Minolta CM-2006d) were determined. The pH was also determined at 1 and 3 hours after slaughter to study the pH fall during ageing, using the negative exponential function as described by Bruce (2004): $pHt= pH_f + ae^{-kt}$ where pHt is the muscle pH at different time, pH_f is final pH, "a" estimates the extent of pH decline and "k" is the exponential constant of pH decline.

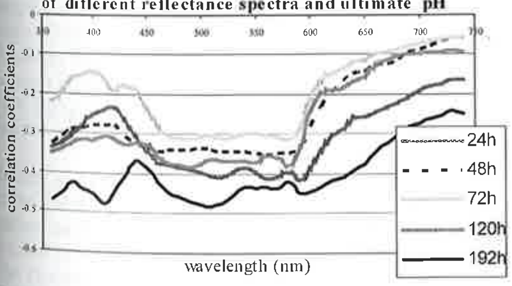
The model was fitted to the pH data of each animal, using proc NLIN procedure (SAS institute, 1999) to obtain respective "a" and "k" coefficients.

The reflectance spectra obtained at different times were correlated with ultimate pH (pHu) to choose the best spectrum to use in multivariate analysis. PLS (Partial Least Square regression) multivariate statistical analysis (Martens and Naes, 1989) is used for creating calibration models for spectroscopic data. The wavelength selection procedure consists of finding the number of variables that give the minimum value of RMSEP (root mean square error of prediction), found by cross-validation. Before performing the PLS regression, spectral outliers were identified and eliminated. The multivariate data analysis was performed with a chemometric program the Unscrambler 9.1 (CAMO, Trondheim, Norway).

Results and Discussion

The corresponding Pearson correlation coefficients between the reference pH and visual spectra (360nm to 740nm) are shown in Figure 1; the maximum correlation coefficients were achieved at wavelength interval between 460nm and 640nm, Soret peak at 192h visual spectrum and between 450 and 580nm at 24h visual spectrum, while the other ones showed a pronounced decrease in a narrow spectral range around 420nm, coinciding with the Soret peak as reported by Andersen *et al.* (1999). The correlation coefficient between ultimate pH and the single wavelength highlights the possibility of estimating the ultimate pH from a spectrum performed at 24 hours, while the spectra obtained at 48 and 72 hours showed a higher variability. Particularly interesting was the possibility of associating spectrometrics with pH decline over time. PLS analysis (Table 1) evidenced an optimal RMSEP between spectrum data and coefficient of pH decay proposed by Bruce (2004); although a low value of RMSEP, the cross validation didn't show a good index of correlation and slope among predicted and measured pH values. Therefore, with these errors it is necessary to consider

Figure1 Correlation between single wavelength of different reflectance spectra and ultimate pH



also the low capacity of the exponential equation to take account of anomalous courses of pH registered in some animals.

The cross validation between ultimate pH, coefficient "k" and spectrum data at 192 hours however resulted better then the other, in fact a good correlation index and slope was obtained. Moreover, for a good pH and decay prediction it was necessary to eliminate more sample outliers from the spectrum at 24 hours than the other one. The regression coefficients (Figure 2) evidenced principal points to predict the data, in particular the absorbances at the beginning and at the end of spectrum (360, 370nm and from 630 to 740nm), and at the absorption points of myoglobin (470, 580 nm), in particular about six factors of PLS were necessary to estimate the pHu in validation phase. Also if it was possible to estimate the ultimate pH from the visible spectrum of reflectance obtained both at 24 and at 192 hours. Chianini meat gave more problems: this in fact showed limited pH decay and some samples had maintained high pH also after eight days of ageing, in fact it had a greater frequency of outlier values (Table 2). Considering the relationship between predicted and measured pH, Chianina values were undervalued, while Marchigiana and Piemontese pH values were slightly overvalued.

The results obtained indicate that the use of a spectroscopic method for measuring pH may be possible, improving determination rapidity, while further studies are necessary to obtain from the spectrometric data information on the pH trend and an estimation of pH decline after rigor.

Table 1: Cross-validation results for prediction pH with visible spectrometer.

	Spectrum at 24 h		Spectrum at 192h	
	pHu	"k"	pHu	"k"
N outliers	8	15	4	9
PLS factors	7	6	6	6
RMSEP	0.0346	0.0503	0.0422	0.0417
Correlation index	0.79	0.64	0.81	0.74
Slope	0.694	0.56	0.741	0.62

Figure 2. Estimated regression coefficient for predicting pHu by spectrum at 192 hours

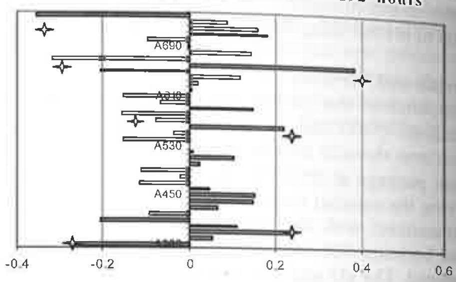


Fig.3 Predicted versus measured pH values

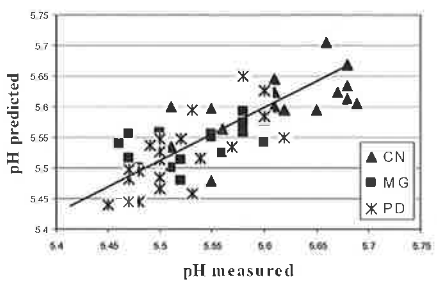


Table 2: pH and exponential decay constant "k" in the different breed.

	pH at 3hours	Ultimate pH	"k"
CN	6.20	5.62a	0.125
MG	6.24	5.53b	0.183
PD	6.21	5.51b	0.196
RMSE	0.073	0.051	0.092

different letters mean significant differences (p< 0.05).

References

Andersen J.R., Borggaard C., Rasmussen A.J., Houmoller L.P., (1999). Meat Science 53: 135-141.

Bendall, J. R. (1979). Meat Science, 3: 143–157.

Bruce, H.L., (2004) Meat Science 66: 507-512.

Devine C.E, and Graafhuis A.E., (1995). Meat Science 39: 285.

Martens, H., and Martens, M. (2001). London, UK: Wiley.

Swatland, H. J., (1989). Canadian Institute Food Science and Technology Journal, 22: 390–402.

Tompson J., (2002). Proceeding of the 48th International Congress of Meat Science and Technology, 1: 17-28.