PRINCIPAL COMPONENT REGRESSION OF NEAR–INFRARED REFLECTANCE SPECTRA FOR BEEF TENDERNESS PREDICTION

B. Park, Y. R. Chen, W. R. Hruschka, S. D. Shackelford, M. Koohmaraie

ABSTRACT. *Tenderness is the most important factor affecting consumer perception of eating quality of meat. In this paper, the development of the principal component regression (PCR) models to relate near–infrared (NIR) reflectance spectra of raw meat to Warner–Bratzler (WB) shear force measurement of cooked meat was presented. NIR reflectance spectra with wavelengths from 1100 to 2498 nm were collected on 119 longissimus dorsi meat cuts. The 1st principal component (or factor) from the absorption spectra log(1/R) showed that the most significant variance from the spectra of tough and tender meats were due to the absorptions of fat at 1212, 1722, and 2306 nm and water at 1910 nm. The distinctive fat absorption peaks at 1212, 1722, 1760, and 2306 nm were found in the 2nd factor of the second derivative spectra of meat. In addition, the local minima in the 2nd principal component of the second derivative spectra showed the importance of water absorption at 1153 nm and protein absorption at 1240, 1385, and 1690 nm. When the absorption spectra between 1100 nm and 2498 nm were used, the coefficient of determination (R^2) of the PCR model to predict WB shear force tenderness was 0.692. The R^2 was 0.612 when the spectra between 1100 nm and 1350 nm were analyzed. When the second derivatives of the spectral data were used, the R^2 of the PCR model to predict WB shear force of the meat was 0.633 for the full spectral range of 1100 to 2498 nm and 0.616 for the spectral range of 1100 to 1350 nm.*

Keywords. *Beef tenderness, Quality, Reflectance, NIR spectrophotometry, Principal component analysis (PCA).*

Among the many quality factors of meat, such as texture, flavor, juiciness, appearance, and aroma, the texture or tenderness in particular is considered the most important in determining the meat eating quality (Morgan et al., 1991; Koohmaraie et al., 1995; Miller et al., 1995). Inconsistency in meat tenderness has been identified as one of the major problems facing the beef industry (Morgan et al., 1991; Savell and Cross, 1992). Because consumers consider tenderness to be the major determinant of eating quality of meat, it is important to develop techniques to objectively predict meat tenderness to supplement or replace the current USDA quality grading system. Even though tenderness is considered to be the major determinant of meat quality, no rapid method exists for the grader or retailer to use to determine tenderness of meat. The current tenderness measurement by taste panels is a subjective method, and is a time–consuming process, because it requires long sample preparation time. The Warner–Bratzler (WB) shear device is widely used in the United States for measuring tenderness of cooked meat. Although it is an objective method, it is also time consuming and destructive. Thus, an objective, nondestructive, and rapid technique for assessing beef tenderness needs to be developed.

Near–infrared (NIR) spectroscopy has become an important tool to measure chemical composition and moisture content of meat and meat products. Ben–Gera and Norris (1968) investigated NIR transmittance for measuring fat and moisture contents in emulsions of meat products. Later, NIR spectroscopy was used to measure moisture and biochemical properties such as fat and protein in emulsified lamb, pork, and beef (Kruggel et al., 1981; Iwamoto et al., 1981; Lanza, 1983). These studies, however, were carried out on ground or emulsified meats. NIR spectroscopy has also been applied to the measurement of chemical composition and textural attributes of raw and cooked meat. The composition (moisture, fat, and protein) has been measured for raw poultry (Renden et al., 1986; Valdes and Summers, 1986). Recently, Marks and Chen (1996) evaluated cooked ground poultry patties using NIR techniques. NIR spectroscopy was also applied to predict total pigment values by measuring optical density of raw fresh meat (Mitsumoto et al., 1991). NIR reflectance and interactance measurements were used to classify wholesome and unwholesome carcasses based on myoglobin measurement, which affects pigment content of poultry meat (Chen and Massie, 1993; Chen et al., 1994; Chen et al., 1996a; Chen et al., 1996b). Changes in NIR spectra of beef muscles during conditioning and aging of beef were investigated, and the feasibility of NIR spectroscopy in

Article was submitted for review in May 1998; approved for publication by the Food & Process Engineering Institute of ASAE in January 2001.

Mention of any company or trade name is for the purpose of description only and does not imply endorsement of the products by the USDA.

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the prediction and assessment of meat sensory attributes was reported (Hildrum et al., 1994). NIR technology has the potential to be used for assessing the tenderness of meat (Park et al., 1998a; Park and Chen, 1998b).

Although NIR spectroscopy has been demonstrated to be a promising method for assessing meat quality of individual carcasses, more research should be continued to develop systems that are accurate, reliable, practical, low cost, and rapid, and that can be readily adopted by the meat industry, especially for tenderness measurement. The objective of this study is to develop a technique to use near–infrared (NIR) spectroscopy of raw meat to predict its tenderness rapidly and nondestructively. More specifically, the objectives are: 1) to measure NIR reflectance on raw meat and 2) to develop models to relate NIR reflectance spectra of raw meat to WB shear force measurement of cooked meat, using the principal component regression (PCR) models.

Materials and Methods

Materials

Meat samples of longissimus dorsi (LD) muscle from 119 beef carcasses were used for establishing NIR spectra measurement. Approximately 25–mm thick steaks were excised from the LD muscles of the thirteenth rib from the right side of each carcass. The samples were vacuum–packed in polyethylene bags and frozen and stored at –30° C. The samples were completely thawed for 24 h at 2° C before NIR spectra were collected. From each steak, two cylindrical shaped samples of approximately 38–mm diameter were excised using a stainless steel punch force corer that allowed the meat sample to fit in a quartz window–clad cylindrical cell. In the sampling procedure, excessive fat and connective tissue were avoided to minimize sampling errors. Each sample was cut to make three or four (the number was dependent on the size of ribeye muscle) circular slices of 8 mm thickness from the cylindrical pieces of meat. A total of 405 disk cut meat samples were used to collect NIR reflectance spectral data.

Shear Force Measurement

Steaks were cooked on an electric grill to an internal temperature of 70°C. Copper–constantan thermocouples were placed in the geometric center of each steak and temperature was monitored. For shear force measurement, cooked steaks were cooled for 24 hours at 4°C before removal of six cores (1.27 cm in diameter) parallel to the longitudinal orientation of the muscle fibers. Each core was sheared once with a Warner–Bratzler attachment using an Instron universal testing machine (Canton, Mass.). The cross–head speed was set at 20 cm/min. The averages of the maximum force readings were used for data analysis as a reference to develop prediction models for meat tenderness by NIR measurement.

NIR Reflectance Measurement

A scanning monochromator (model 6500, NIR Systems, Silver Spring, Md.) was used to collect reflectance (R) readings over a wavelength range of 1100 – 2498 nm in 2 nm increments, yielding 700 values per spectrum. Two pairs of lead sulfide detectors collected the reflectance spectra. The absorbance spectrum, recorded as log(1/R) for each meat sample, was gathered on a spectrophotometer equipped with a rotating drawer. Reflected energy readings were referenced to corresponding readings from a ceramic disk. A reference scan was collected and stored to computer memory before each sample was scanned. The spectra from three or four circular slices of each sample were averaged to produce one spectrum per sample for the development of chemometric models to predict meat tenderness. The spectrum of a meat sample was the average of 32 successive scans (i.e. grating oscillations), altogether taking approximately 20 seconds per slice.

Principal Component Analysis

Principal component analysis (PCA) or spectral decomposition produces a reduced representation of the training data based on the maximum variations between the spectra. This produces a small set of defined numbers that can be used for discrimination, since it provides an accurate description of the entire training set. Effectively, PCA finds a set of mathematical spectra (or factors) that contains the maximum variations common to all spectra in a data set. This sets up a new space where each spectrum in the original group of data can be modeled by a linear combination of these factors (Chen et al., 1997). The linear combination coefficients or scores, which determine how much of each factor is needed to reconstruct the original spectrum, can be calculated from this set of factors and the original data. Each spectrum will have its own unique set of scores; therefore, a spectrum can be represented by its PCA scores in the factor space instead of intensities in the wavelength space.

Principal Component Regression Model

The mean spectrum was first calculated from all of the calibration spectra and then subtracted from every calibration spectrum. Mean centering would enhance the subtle differences between the spectra. Since eigenvector methods calculate the principal components based on changes in the absorption data, the ability of the calculation to detect the differences between the calibration spectra would improve the model. When the PCA algorithm has processed the training data, it is reduced to two main matrices; the eigenvectors and the scores. The matrix expression of the PCR model for the spectral data can be obtained by equation 1:

$$A = SV + E_A \quad (1)$$

where

A = spectral absorption matrix (n by w)
S = score values matrix (n by m)
V = eigenvector matrix (m by w)
E_A = residual spectra matrix (n by w)

and

n = number of spectra
w = number of wavelengths

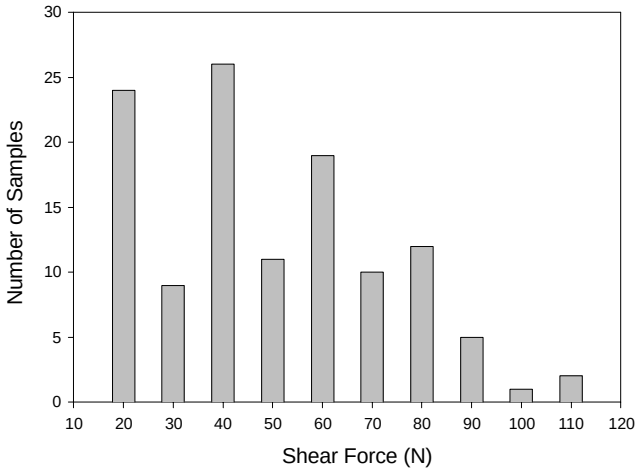

Figure 1. Sample distribution of longissimus muscle based on shear force measurement for principal component regression (PCR) model.

m = number of principal component eigenvectors.

As the scores in the S matrix are calculated from each spectrum, and a spectrum is represented by a collection of absorption at a series of wavelengths, it is possible to obtain regression model for concentrations against the score matrix as equation 2:

$$Y = CS' + E_Y \quad (2)$$

where

Y = constituent concentrations matrix (p by n)

C = regression coefficients matrix (p by m)

E_Y = error matrix (p by n) and

p = number of constituents for calibration

Prime indicates the transpose of the matrix.

As with least square regression, the coefficients matrix can be solved by the regression equation:

$$C = YS(S'S)^{-1} \quad (3)$$

From equation 1, the score matrix of the spectra can be obtained by equation 4 after eliminating the noise error matrix:

$$S = AV^{-1} = AV' \quad (4)$$

For equation 4, the transpose matrix of V can substitute its inverse matrix because V matrix of eigenvector is an orthonormal matrix. By combining the concentration (eq. 2) with the score (eq. 4), the PCR equation can be obtained as equation 5:

$$Y = CVA' + E_Y \quad (5)$$

As described above, the PCR is a two–step process. The PCA eigenvectors and scores, which represent the largest common variations among all the spectra in the calibration data, are calculated first, and then the prediction model is developed for scores against the constituent concentrations using a regression method. A PCR model should be built by performing a selection on the scores to determine which factors should be used to build a model for each constituent.

Results and Discussion

The meat samples were excised from the longissimus muscle of 119 beef animals, with tenderness ranging from

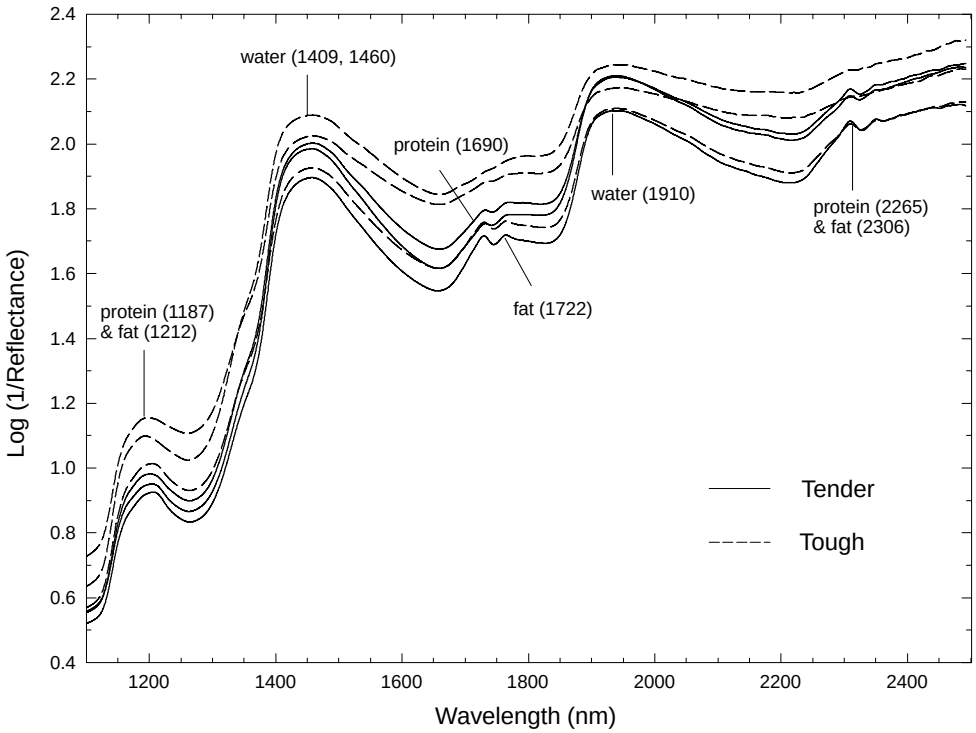

Figure 2. Near–infrared reflectance spectra of frozen and thawed longissimus muscles for tenderness measurement.

19.7 N to 114.7 N (measured in WB shear force) (fig. 1). The mean WB shear force of the samples and its standard deviation were 53.5 N and 21.7 N, respectively. For the shear force prediction, the coefficients of determination (R^2) of the best PCR models were determined by applying the cross–validation procedure.

NIR Reflectance Characteristics of Longissimus Muscle

Figure 2 show that the tough meat (shear force = 114.7 N) had a higher absorption than the tender meat (shear force = 37.3 N) at most wavelengths, particularly for the wavelengths between 1100 and 1350 nm. This is similar to the result of a previous report by Hildrum et al. (1994). Obvious absorption differences existed between tough and tender meats at protein absorption bands at 1187, 1690, and 2265 nm; fat absorption bands at 1212, 1722, and 2306 nm; and water absorption bands at 1409, 1460, and 1910 nm, respectively. Significant variations in spectra among samples from the same steak were also found. This showed that a gradient in tenderness exists within the longissimus muscle and proved that a tenderness gradient also exists within a steak obtained from the longissimus muscle (Alsmeyer et al., 1965; Sharrah et al, 1965; Smith et al., 1969). In our model development, because the tenderness of each steak was represented by a WB shear force value for the steak, the spectra of the disk samples sliced from each steak were averaged and used for the calibration and validation to minimize variation within the samples.

Figure 3 shows the comparison of the absorbance between tender (37.3 – 42.2 N) and tough (99.1 – 114.7 N) meats and their second derivatives. As shown in the bottom graph, the absorption values for tough meats were higher than those of tender meats at all wavelengths. The peaks of the difference curve were mostly at the protein and fat bands. The smallest differences between tender and tough meats were found at the water bands (1460 and 1910 nm).

To improve the performance of identification of spectra between tender and tough meat, the second derivative of each spectrum was calculated and compared (Fig. 3, top). The second derivatives of NIR spectra help resolution of overlapping peaks and removal of baseline variations (Hruschka, 1987). The wavelength bands, which occurred at the transition from maximum to minimum or the vice versa of the second derivatives of the spectra, were 1380 and 1870 nm. This implies that no spectral absorption difference was found between tender and tough meat samples at these bands.

Principal Component Analysis for Meat Tenderness

Principal component analysis (PCA) was used to reduce the contribution of noise in the modeling procedure. Absorption and second derivative spectra were used to correlate with WB shear force values of the meat. Figure 4 shows the first three principal components extracted from the calibration data set of the absorption spectra. For the 1st principal component (or the first factor) from the absorption spectra, the most significant variance from the spectra of tough and tender meats was due to absorptions of fat at 1212,

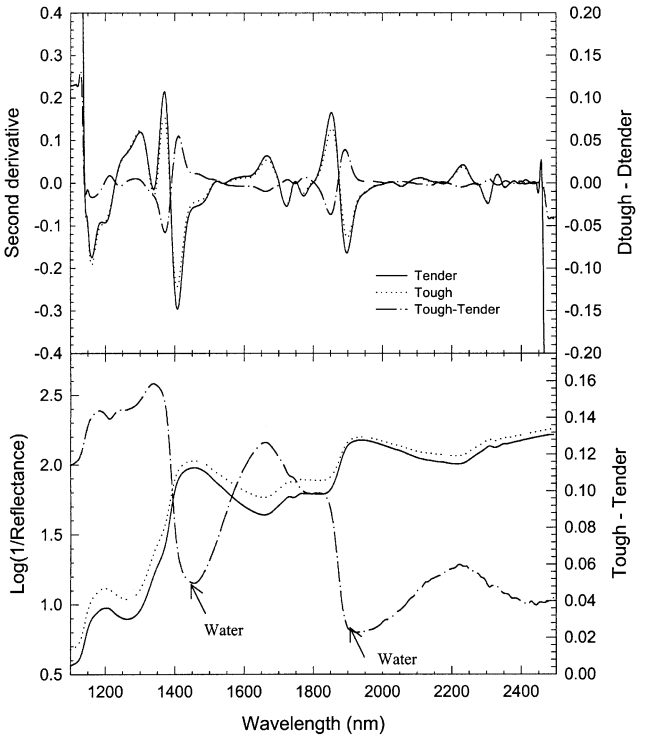

Figure 3. Absorption of near–infrared spectra and their second derivatives of tender and tough meat samples.

1722, and 2306 nm; protein at 1187 nm; and water at 1910 nm. In the 2nd factor, the peaks of absorption were also found at 1458 nm for protein and 1460 and 1910 nm for water. As shown in the 3rd factor of figure 4, the high absorption bands of fat were found obviously at the wavelengths of 1212, 1722, 1760, and 2306 nm. In addition, high absorption peaks were observed at 1910 and 2345 nm of water and protein, respectively.

Figure 5 illustrates the absorption wavelength bands of the second derivative spectra of the longissimus dorsi meat cuts. It shows that the major contribution to the variance among the tough and tender meats was from fat absorptions. For the 1st principal component, the peaks occurred at the fat absorptions at 1212, 1722, and 2306 nm, and the protein absorptions at 1187, 1365, and 2345 nm. The distinctive fat absorption peaks at 1212, 1722, 1760, and 2306 nm occurred in the 2nd factor. The water absorption at 1900 nm, the fat or protein absorption at 2265 nm, and the protein absorption at 2345 nm were also presented in the 2nd factor of the second derivative spectra of the meat. The valleys in the 2nd factor of the second derivative spectra indicated the importance of water absorption at 1153 nm and protein absorption at 1240, 1385, and 1690 nm, even if these peaks were not significant compared with the wavelength bands of fat absorption. For the 3rd factor, the absorption bands for fat and protein were significant at 1212, 1722, 1760, and 2380 for fat and 1240, 1690, and 2345 nm for protein, respectively.

PCR Model for Meat Tenderness Prediction

Based on the absorption spectra, log(1/R) and their second derivatives, principal component regression (PCR) models for predicting tenderness (WB shear force) were developed.

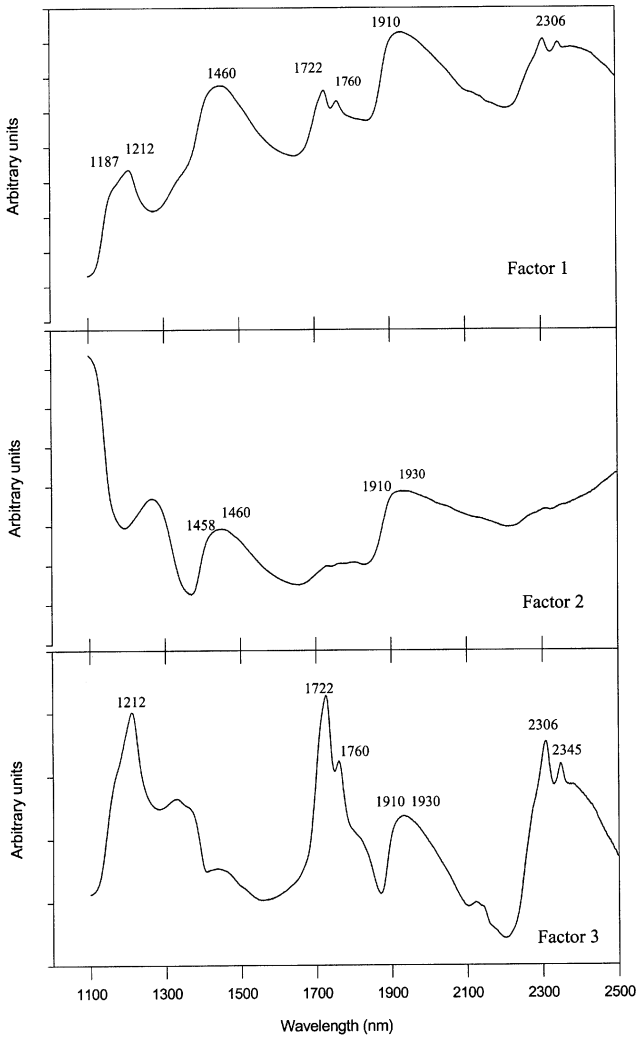

Figure 4. Principal component factors extracted from the calibration data set of the NIR spectra collected from longissimus muscle for tenderness measurement.

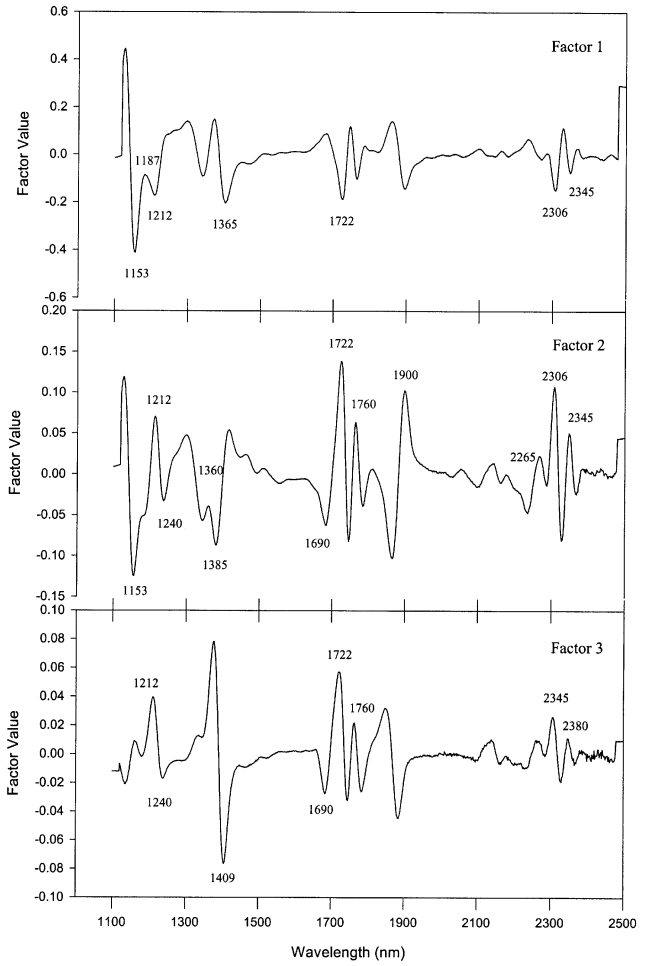

Figure 5. Principal component factors extracted from the calibration data set of the second derivative NIR spectra collected from longissimus muscle for tenderness measurement.

Ranges of wavelengths were selected for the models. When the absorption spectra between 1100 nm and 2498 nm were used, the coefficient of determination (R^2) of the PCR model to predict WB shear force tenderness was 0.692. When the spectra between 1100 nm and 1350 nm were analyzed, the R^2 was 0.612.

The R^2 was 0.535 when four wavelength bands of 1567 to 1617 nm, 1663 to 1713 nm, 1829 to 1879 nm, and 2115 to 2165 nm (selected from the multivariate data analysis) were used (fig. 6). When the second derivatives of the spectra were used for the PCR models to predict the WB shear force for meat tenderness, the R^2 was slightly better than the model with absorption spectra between 1100 and 1350 nm wavelength bands. When the full spectral range of 1100 to 2498 nm was used, the R^2 of the PCR model to predict WB shear force of the meat was 0.633. The R^2 decreased to 0.616 when the second derivatives of the spectra of wavelengths between 1100 and 1350 nm were selected (fig. 7).

The R^2 value (0.69) of the PCR model was observed higher than that of the partial least squares (PLS) model (R^2 = 0.67) or multiple linear regression (MLR) model (R^2=0.63) published previously (Park et al., 1998a). Moreover, based on the different models, the results from the NIR measurement for predicting tenderness were consistent. Even though the PCR model could not be implemented in the beef industry to predict tenderness, refinement of this model could be applied for classifying tenderness, such as tough, intermediate, tender, with high accuracy using nondestructive NIR measurement.

SUMMARY AND CONCLUSIONS

An objective, nondestructive, and rapid technique for assessing beef tenderness needs to be developed. The principal component regression (PCR) technique was utilized to determine cooked meat tenderness using NIR reflectance measurement on raw meat. The tough meat (shear force = 114.7 N) had an overall higher absorption than the tender meat (shear force = 37.3 N) at most wavelengths, particularly for the wavelengths between 1100 and 1350 nm. There existed obvious absorption differences between tough and tender meats at protein absorption bands at 1187, 1690, and 2265 nm; fat absorption bands at 1212, 1722, and

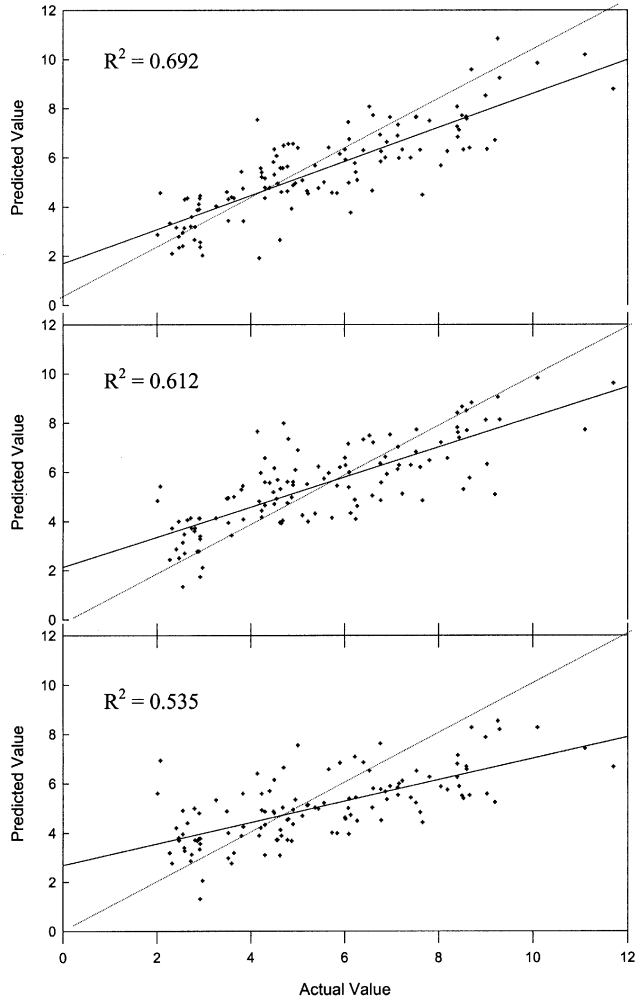

Figure 6. Beef longissimus muscle tenderness prediction by NIR spectroscopy using PCR model for the wavelength of (a) 1100 – 2498 nm; (b) 1100 – 1350 nm; (c) four selected bands: 1567 –1617 nm, 1663 – 1713 nm, 1829 – 1879 nm, and 2115 – 2165 nm.

2306 nm; and water absorption bands at 1409, 1460, and 1910 nm.

For the 1st principal component from the absorption spectra, it can be seen that the most significant variance from the spectra of tough and lean meats was due to the fat absorptions at 1212, 1722, and 2306 nm and water absorption at 1930 nm. For the 2nd principal component of the second derivative spectra of the meat, the distinctive absorption peaks due to fat absorption at 1212, 1722, 1760, and 2306 nm; water at 1900 nm; fat or protein at 2265 nm; and protein at 2345 nm were also presented. In this factor, significant absorption was found for water at 1153 nm and protein at 1240, 1385, and 1690 nm.

Based on the absorption spectra, log(1/R), the coefficient of determination (R^2) of the principal component regression (PCR) model to predict Warner–Bratzler (WB) shear force tenderness was 0.692 when the absorption spectra between 1100 nm and 2498 nm were used. In case of the second derivatives of the spectra, the R^2 was 0.633 when the full spectral range of 1100 to 2498 nm was used. Finally, in this study, the biochemical constituent composition of fat and

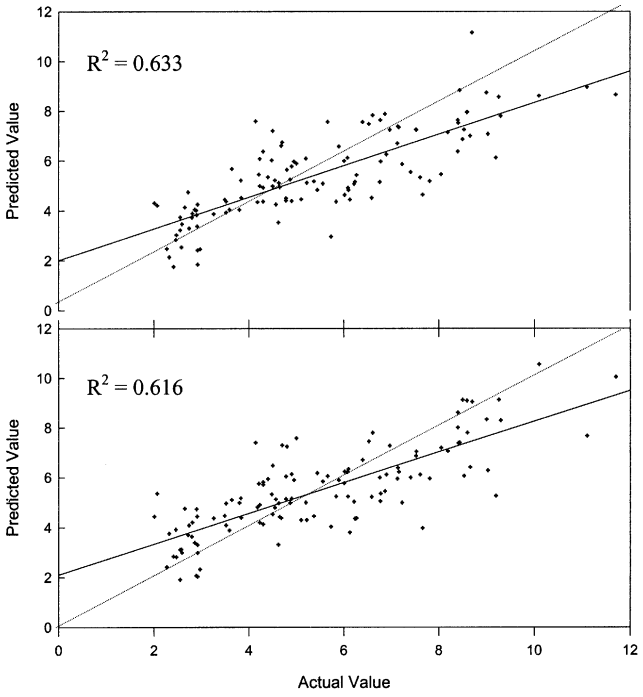

Figure 7. Beef longissimus muscle tenderness prediction by NIR spectroscopy using PCR model for the second derivative of wavelengths (a) 1100 – 2498 nm and (b) 1100 – 1350 nm.

protein were identified as absorbers of NIR spectra. Other factors that affect meat tenderness, such as collagen and the amount of connective tissue, should be studied for better understanding of how those parameters would be correlated with the absorption of NIR spectra.

ACKNOWLEDGEMENTS

The authors thank to Dr. S. B. Lee, a visiting scientist at the Instrumentation and Sensing Laboratory, who helped with sample preparation for these experiments.

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