

Near-Infrared Reflectance Analysis for Predicting Beef Longissimus Tenderness¹

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ABSTRACT: Near-infrared reflectance spectra (1,100 to 2,498 nm) were collected on beef longissimus thoracis steaks for the purpose of establishing the feasibility of predicting meat tenderness by spectroscopy. Partial least squares (PLS) analysis (up to 20 factors) and multiple linear regression (MLR) were used to predict cooked longissimus Warner-Bratzler shear (WBS) force values from spectra of steaks from 119 beef carcasses. Modeling used the combination of $\log(1/R)$ and its second derivative. Overall, absorption was higher for extremely tough steaks than for tender steaks. This was particularly true at wavelengths between 1,100 and 1,350 nm. For PLS regression, optimal model conditions ($R^2 = .67$; SEC = 1.2 kg) occurred with six PLS factors. When the PLS model was tested against the validation

subset, similar performance was obtained ($R^2 = .63$; SEP = 1.3 kg) and bias was small (<.3 kg). Among the 39 samples in the validation data set, 48.7, 87.7, and 97.4% of the samples were predicted within 1.0, 2.0, and 3.0 kg, respectively, of the observed Warner-Bratzler shear force value. The optimal PLS model was able to predict whether a steak would have a Warner-Bratzler shear force value < 6 kg with 75% accuracy. The R^2 of MLR model was .67, and 89% of samples were correctly classified (< 6 vs > 6 kg) for Warner-Bratzler shear force. These data indicate that NIR is capable of predicting Warner-Bratzler shear force values of longissimus steaks. Refinement of this technique may allow nondestructive measurement of beef longissimus at the processing plant level.

Key Words: Beef, Tenderness, Reflectance, Spectrophotometry, Statistical Analysis, Regression

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Introduction

Inconsistency in meat tenderness has been identified as one of the major problems facing the beef industry (Smith et al., 1995). One method to limit the consequences of inadequate tenderness is to objectively evaluate tenderness and market cuts appropriately. Technology to accurately classify beef for tenderness based on cooked longissimus shear force has been developed (Shackelford et al., 1997 and unpublished data). Some beef processors are implementing that technology, but others have been reluctant to implement that technology because it is

destructive. If possible, it is preferable to develop an accurate, nondestructive method to objectively predict meat tenderness.

Hildrum et al. (1994) reported that the near-infrared reflectance (NIR) spectra of beef muscles changed during aging. Given that a variation in the rate of aging causes most of the variation in tenderness of longissimus steaks from the carcasses of young, grain-fed cattle (Whipple et al., 1990; Shackelford et al., 1991), NIR spectroscopy may be able to predict variation in tenderness of longissimus steaks.

The objective of this study was to determine the relationship between NIR spectra and Warner-Bratzler shear force, an objective measurement of meat tenderness, of beef longissimus steaks.

Materials and Methods

Materials. Carcasses (n = 119) used in this experiment were selected based on Warner-Bratzler shear force from a population containing over 600 carcasses. These carcasses were selected based on longissimus Warner-Bratzler shear force to maximize

¹Names are necessary to report factually on available data; however, the USDA neither guarantees nor warrants the standard of the product, and the use of the name by USDA implies no approval of the product to the exclusion of other products that may also be suitable. The authors are grateful to Carol Grummert for her secretarial assistance.

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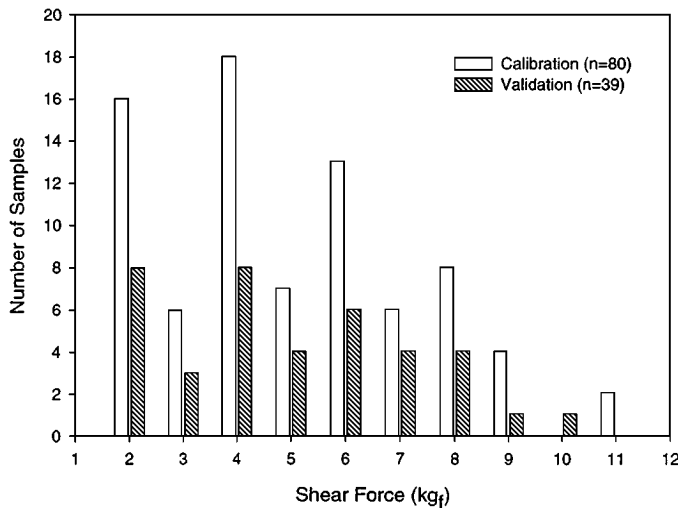


Figure 1. Distribution of longissimus Warner-Bratzler shear force values of samples used for partial least squares (PLS) model calibration and validation.

variation in shear force, and, therefore, they do not represent a typical distribution of shear values (Figure 1). For this experiment, two longissimus thoracis steaks (approximately 2.5 cm thick) were obtained from each carcass and were frozen (-30°C) at either 7 (n = 69) or 14 (n = 50) d after slaughter. One steak from each carcass was used to measure cooked longissimus Warner-Bratzler shear force. The other steak was used for NIR spectroscopy.

Warner-Bratzler Shear Force. Steaks were thawed for 24 h at 4°C , broiled to an internal temperature of 40°C , turned, and broiled to a final internal temperature of 70°C . Steaks were placed in bags and cooled for 24 h at 4°C before removal of six cores (diameter = 1.27 cm) parallel to the longitudinal orientation of the muscle fibers. Each core was sheared once perpendicular to the longitudinal orientation of the muscle fibers with a Warner-Bratzler shear attachment using an Instron Universal Testing Machine (Instron Corp., Canton, MA).

NIR Spectroscopy. Steaks were completely thawed for 24 h at 2°C before NIR spectra were gathered. To obtain a sample that would fit into the spectrophotometer's quartz window-clad cylindrical cell, two cylindrical cores of 38-mm diameter were excised from each steak using a punch force corer. Depending on the thickness of the steak, one or two 8-mm-thick circular slices were obtained from each core. A total of three or four (average = 3.4) slices were obtained from each steak.

A scanning monochromator (model 6500, NIR-Systems, Silver Spring, MD) was used to collect reflectance (**R**) readings over a wavelength range of 1,100 to 2,498 nm in 2-nm increments, yielding 700 values per spectrum. Two pairs of lead sulfide detectors collected the reflectance spectra. The absor-

bance spectrum recorded as $\log(1/R)$ for each meat disk 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 spectrum of each slice was the average of 32 successive scans (i.e., grating oscillations), which altogether took approximately 20 s per slice. The spectra from the three or four circular slices of each steak were averaged to produce one spectrum per steak for the development of chemometric models to predict meat tenderness.

Chemometric Analyses

The spectra of the 119 steaks were divided into a calibration subset (80 steaks) and a validation subset (39 steaks). The methods of partial least squares (**PLS**) and multiple linear regression (**MLR**) were used for all chemometric models. The MLR procedure on spectral data is described in Hruschka (1987), and the mathematics of PLS are well developed and described in Lindberg et al. (1983).

Partial Least Squares Model. The PLS procedure was applied directly to the $\log(1/R)$ spectra with the wavelength region 1,100 to 2,498 nm. Cross-validation was performed during model development. For cross-validation, each sample was temporarily removed from the calibration data set, one sample at a time. A mean-centering data processing algorithm was applied to the calibration model. On completion of calibration, the model was applied to the validation data set. Model performance was reported as the standard error of calibration (**SEC**), the coefficient of determination (R^2), the standard error of prediction (**SEP**), and the average difference between values predicted by the model and observed values (bias) and each term was calculated on the validation set. For the calibration performance, the SEC was calculated as

$$\text{SEC} = \sqrt{\frac{\sum_{i=1}^n (Y_i - R_i)^2}{n - f - 1}} \quad [1]$$

where Y_i and R_i are the predicted and observed shear force values for sample i , respectively. The value of n is the number of samples, and f is the number of factors in the model. For the model performance, the SEP was calculated as

$$\text{SEP} = \sqrt{\frac{\sum_{i=1}^n [(Y_i - R_i) - \bar{D}]^2}{n - 1}} \quad [2]$$

where \bar{D} is the average of $(Y_i - R_i)$, also referred to as *bias*.

Multiple Linear Regression Model. For the spectral pretreatment of the MLR procedure, mean smoothing was conducted with three-point averaging (Hruschka, 1987) followed by $\log(1/R)$ transformation and computation of second derivatives. The MLR procedure consisted of a stepwise search for the best combination of wavelengths in the equation:

$$\text{Warner-Bratzler shear force} = k_0 + k_1 \cdot f(\lambda_1) + k_2 \cdot \{f(\lambda_2) - f(\lambda_3)\} + k_3 \cdot \Delta^2 f_g(\lambda_4) \quad [3]$$

where $f(\lambda_1)$ is the $\log(1/R)$ spectrum and the k_i are constants. Second derivatives were computed as

$$\Delta^2 f_g(\lambda_i) = \frac{f(\lambda_i - g) - 2f(\lambda_i) + f(\lambda_i + g)}{g^2} \quad [4]$$

where i is the wavelength index at which the second derivative is evaluated and g is the finite difference gap. For the MLR model, four terms that include the first difference and the second derivatives of the spectrum were examined in a stepwise manner that yielded the highest coefficient of determination (R^2). For the development of the classification model, all spectra of steaks with low (<6 kg) Warner-Bratzler shear force values were assigned a constant value of zero, and the spectra of steaks with high (>6 kg) Warner-Bratzler shear force values were assigned a constant value of one.

Results and Discussion

The distribution of meat samples for the calibration and validation sets is shown in Figure 1. By design, these steaks represent a wide range of Warner-Bratzler shear values. The minimum, maximum, mean, and SD of Warner-Bratzler shear force were 2.0, 11.7, 5.5, and 2.2 kg, respectively. The range of values for the validation set fell within the calibration set range. When the best model from the cross-validation procedure was applied to the validation data set, R^2 , SEP, and bias were determined.

NIR Characteristics of Longissimus

Figure 2 shows the NIR spectra $\log(1/R)$ of three longissimus circular slices taken from a single steak having a low (3.8 kg; i.e., tender) Warner-Bratzler shear value and three longissimus circular slices taken from a single steak having an extremely high (11.7 kg; i.e., extremely tough) Warner-Bratzler shear value. Overall, absorption was higher for the circular slices from the extremely tough steak. This was particularly true at wavelengths between 1,100 and 1,350 nm. This finding is similar to the results of Hildrum et al. (1994). There was a large amount of variation in the absorption spectra for the three slices from each steak (Figure 2). Therefore, spectra were

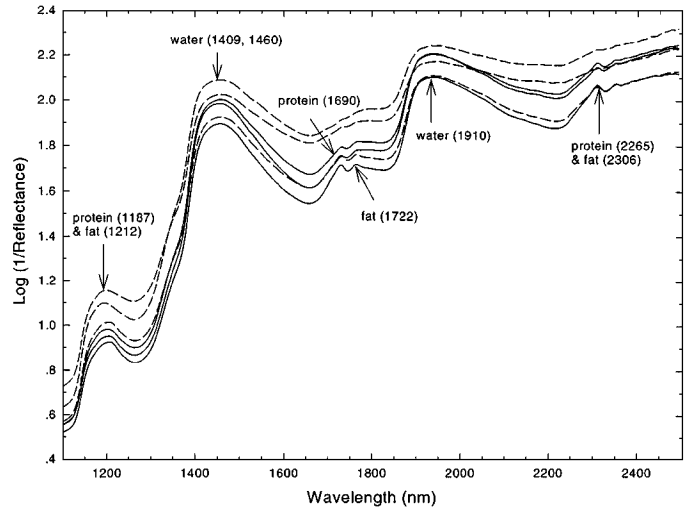


Figure 2. Near-infrared reflectance spectra of three longissimus circular slices taken from a single steak having a low (3.8 kg; i.e., tender; solid line) Warner-Bratzler shear value and three longissimus circular slices taken from a single steak having an extremely high (11.7 kg; i.e., extremely tough; dashed line) Warner-Bratzler shear value.

averaged over all the circular slices from each steak for model development.

Figure 3 shows the mean NIR spectra $\log(1/R)$ for groups of steaks having low (3.8 to 4.3 kg) and extremely high (10.1 to 11.7 kg) Warner-Bratzler shear values. Mean absorption was numerically higher at all wavelengths for the less-tender steaks.

Computation of the second derivatives of NIR spectra allow the resolution of overlapping peaks and removal of baseline variations (Hruschka, 1987). The second derivatives of spectra of tender and tough meat are compared in Figure 3. The most notable transitions (from maximum to minimum or vice versa) of the second derivative of spectra were at 1,380 and 1,870 nm.

PLS Model for Meat Tenderness Prediction

For tenderness prediction, optimal model conditions ($R^2 = .67$; SEC = 1.2 kg) occurred with six PLS factors (Figure 4). When the PLS model was tested against the validation subset (Figure 5), similar performance was obtained ($R^2 = .63$; SEP = 1.3 kg). The NIR predictions were relatively well-clustered about the 45° line (dashed line), on which all points would lie in the case of a perfect model. The optimal PLS model was slightly biased (-.3 kg) when tested against the validation subset.

Among the 39 samples in the validation data set, 48.7, 87.7, and 97.4% of the samples were predicted within 1.0, 2.0, and 3.0 kg, respectively, of the observed Warner-Bratzler shear force value (Figure 6).

Steaks in the validation data set were classified according to observed Warner-Bratzler shear values (< 6 vs > 6 kg), and observed classification was compared with that predicted with the optimal PLS model (Table 1). Of the 23 steaks predicted to have Warner-Bratzler shear values < 6 kg, 83% were observed to have Warner-Bratzler shear values < 6 kg. Of the 16 steaks predicted to have Warner-Bratzler shear values > 6 kg, 75% were observed to have Warner-Bratzler shear values > 6 kg. Therefore, the overall accuracy of the classification was 79%.

MLR Model for Meat Tenderness Classification

For MLR analysis, a wavelength search method (Hruschka, 1987) was used. This search method involves using several terms, each of which could be a value, difference, or quotient of the spectral or derivative data, in the linear regression. The regression used 0 (Warner-Bratzler shear force < 6 kg) or 1 (Warner-Bratzler shear force > 6 kg) as a dependent variable, and threshold values were used for calculating success of classification.

Figure 7 shows the results of the optimal MLR prediction model. The optimal wavelengths selected

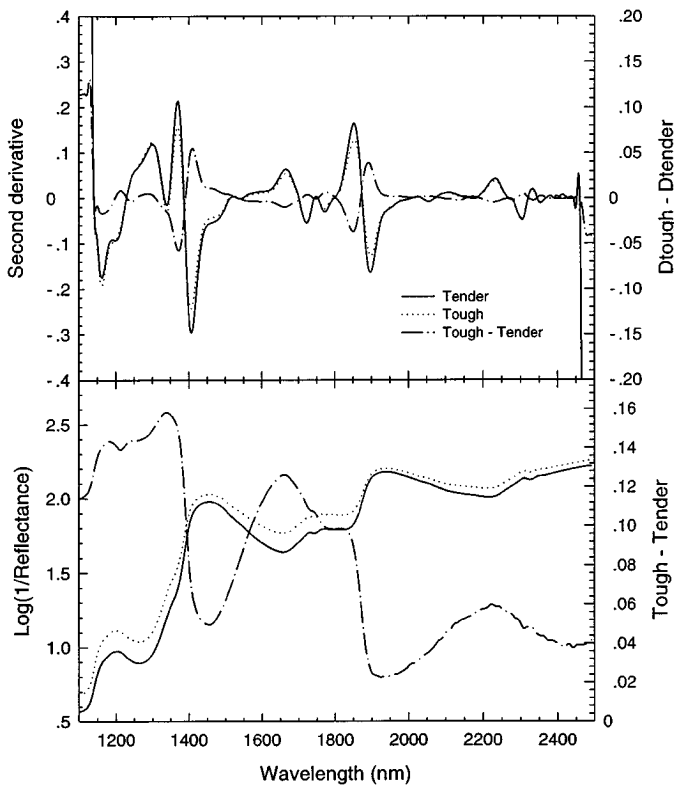


Figure 3. Absorption spectra and their second derivatives for groups of steaks having low (3.8 to 4.3 kg; i.e., tender) and extremely high (10.1 to 11.7 kg; i.e., extremely tough) Warner-Bratzler shear values. Also shown is the difference in reflectance and second derivative between shear force groups.

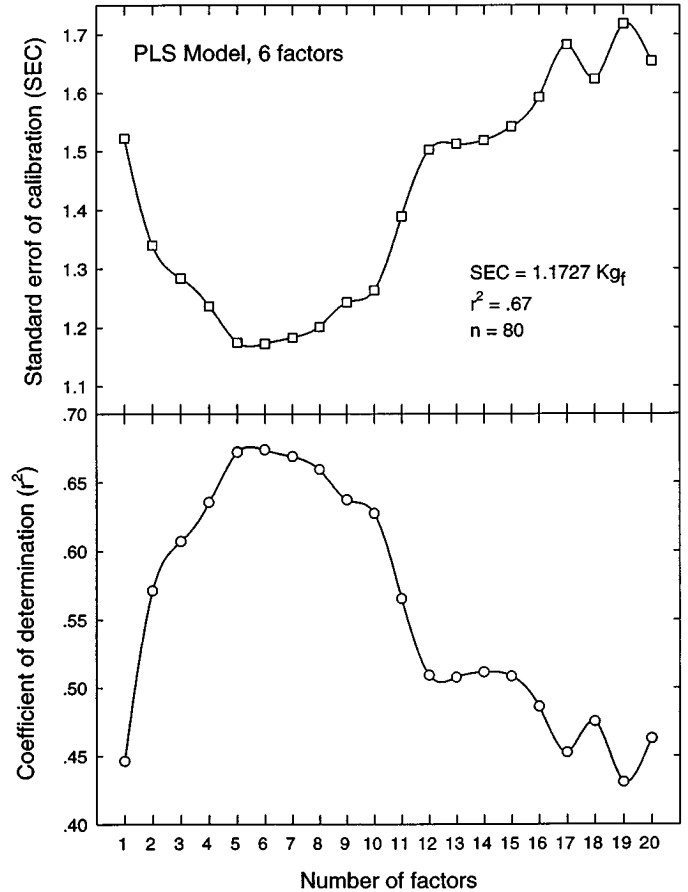


Figure 4. Coefficient of determination and standard error of calibration (SEC) of partial least squares (PLS) model to predict longissimus Warner-Bratzler shear force. The optimal model (SEC = 1.2 kg and R² = .67) contained six partial least squares factors.

for Eq. [3] were $\lambda_1 = 1,854$, $\lambda_2 = 1,688$, $\lambda_3 = 1,592$, and $\lambda_4 = 2,140$ nm when a gap of five points (10 nm) was used. The coefficients of MLR model developed in this experiment were $k_0 = -9.2$, $k_1 = 5.9$, $k_2 = 36$, and $k_3 = 261$ when the optimum wavelengths were used. The R² of MLR model was .67, and 89% of samples were correctly classified (< 6 vs > 6 kg) for Warner-Bratzler shear force.

General Discussion

These data demonstrate that NIR is capable of predicting Warner-Bratzler shear force of longissimus steaks. Probes are currently marketed for convenient measurement of NIR spectra of pharmaceuticals, foods, chemicals, and textiles. If similar results as those obtained in this experiment can be obtained using a commercial probe, then there may be potential to implement this technology in the beef packing industry.

It is unclear whether this technology can be applied to carcasses at packing plants within 1 to 5 d after slaughter and used to accurately predict how tender

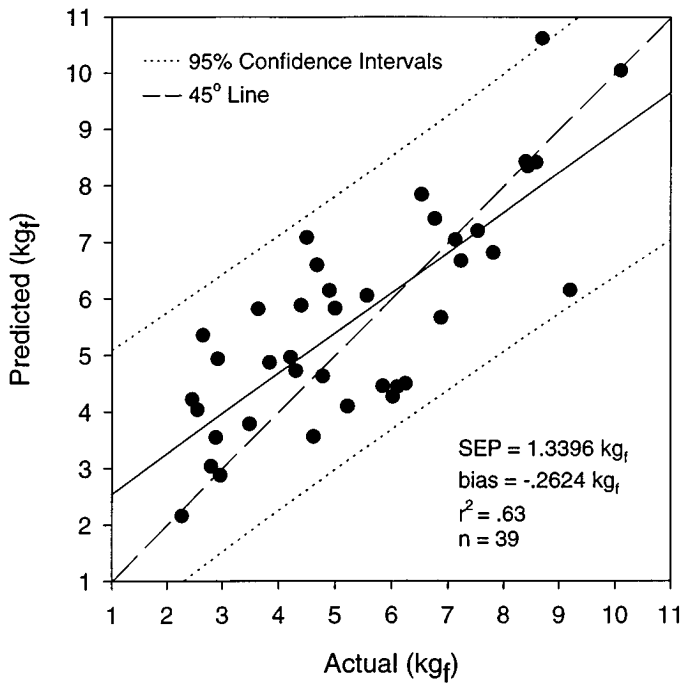


Figure 5. Beef longissimus Warner-Bratzler shear force prediction by near-infrared spectroscopy using partial least squares model. (SEP = standard error of prediction).

the longissimus steaks from those carcasses would be after aging. Given that the majority of variation in Warner-Bratzler shear force of longissimus steaks at 14 d after slaughter can be accounted for by measuring longissimus Warner-Bratzler shear force at 1 d after slaughter (Shackelford et al., 1997), it would

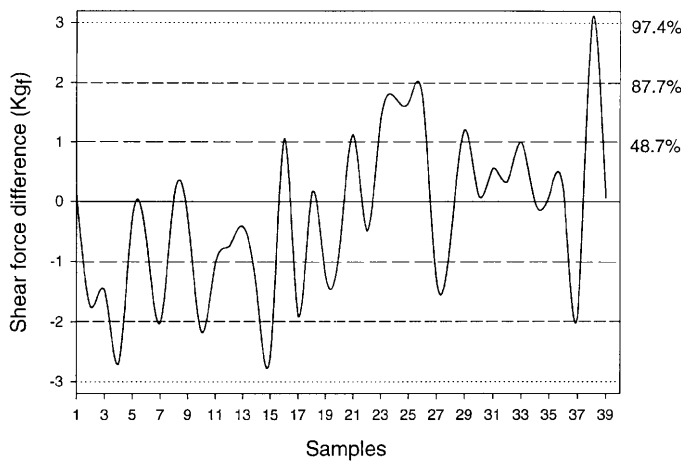


Figure 6. Residuals of predicted shear force values of validation data set by partial least squares model. Among the 39 samples in the validation data set, 48.7, 87.7, and 97.4% of the samples were predicted within 1.0, 2.0, and 3.0 kg, respectively, of the observed Warner-Bratzler shear force value.

Table 1. Accuracy of classifying the validation steaks (n = 39) for Warner-Bratzler shear force using the results of the optimal partial least squares model

| Observed | Predicted | | Type II accuracy, % |
|--------------------|-----------|-------|---------------------|
| | <6 kg | >6 kg | |
| <6 kg | 19 | 4 | 83 |
| >6 kg | 4 | 12 | 75 |
| Type I accuracy, % | 83 | 75 | 79 |

seem that any method capable of accurately predicting longissimus tenderness could be applied at the packing plant level and used to predict the tenderness of longissimus steaks at 14 d after slaughter. However, it should be acknowledged that the level of accuracy obtained would likely be less than what is reported herein because of variability in the aging response. The accuracy of this system for classifying beef for longissimus Warner-Bratzler shear force within a given aging period was similar to the accuracy of classification that Shackelford et al. (1997) reported for classifying longissimus samples for Warner-Bratzler shear force at 14 d after slaughter by measuring Warner-Bratzler shear force at 1 d after slaughter.

Experiments are underway to determine what aspect(s) of meat tenderness can be accounted for by NIR. Information from these experiments may allow repeated measures of those components on the same piece of meat during aging.

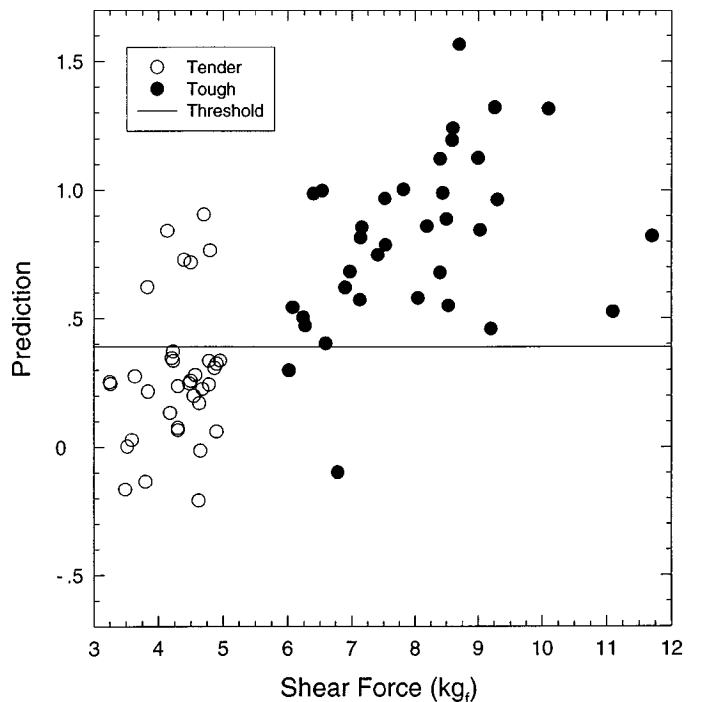


Figure 7. Beef longissimus Warner-Bratzler shear force prediction by near-infrared spectroscopy using the multiple linear regression model ($R^2 = .67$).

Implications

Near-infrared spectroscopy enabled the prediction of beef longissimus Warner-Bratzler shear force. Refinement of this technique may allow nondestructive measurement of beef longissimus at the processing plant level. Therefore, these tools may help facilitate value-based marketing.

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