

¹Department of Animal Science,
Bangladesh Agricultural University,
Mymensingh-2202, Bangladesh.
²Department of Poultry Science,
Bangladesh Agricultural University,
Mymensingh-2202, Bangladesh.

Research Article

Prediction of beef quality traits through mini NIR spectrophotometer and multivariate analyses

MA Hashem^{1*}, SA Tule¹, M Khan¹, MM Rahman¹, MAK Azad¹ and MS Ali²

Abstract

The aim of this study was to test the ability of mini NIR reflectance spectroscopy to predict beef quality traits. Sixty *M. longissimus thoracis* were collected and spectra were obtained prior to beef quality trait analysis. Calibration equations were developed from reference data (n=60) of pH, color traits (lightness, redness and yellowness), drip loss (%), cooking loss (%), CP (%), EE (%), moisture (%), DM (%), and Ash (%) using partial least squares regressions. Predictive ability of the models was assessed by coefficient of determination of cross-validation (R2CV) and root mean square error of cross-validation. Predictions models were satisfactory (R2CV = 0.95) for pH, (R2CV = 0.96) for lightness (L*), (R2CV = 0.96) for redness (a*), (R2CV = 0.97) for yellowness (b*), (R2CV = 0.95) for drip loss, (R2CV = 0.95) for cooking loss, (R2CV = 0.94) for CP, (R2CV = 0.95) for EE, (R2CV = 0.91) for moisture, (R2CV = 0.91) for DM and (R2CV = 0.91) for ash. The ratio performance deviation is 5.35, 5.34, 5.87, 5.16, 4.64, 4.81, 4.45, 4.95, 3.36, 4.73 and 4.47 for L*, a*, b*, pH, drip loss, cooking loss, CP, EE, moisture, DM and Ash respectively which indicates that all values are adequate for analytical purposes. Range error ratio are 20.69, 22.97, 27.11, 18.92, 20.74, 16.20, 17.80, 17.52, 14.96, 17.89 and 17.87 for L*, a*, b*, pH, drip loss, cooking loss, CP, EE, moisture, DM and ash respectively. From the findings of this study it can be concluded that mini NIRS is a suitable tool for a rapid, non-destructive and reliable prediction of beef quality

Introduction

Meat contains high biological important macro and micronutrients, all of which are essential for better human health throughout life. World trade in meat and meat products is forecast at 36.0 million tones in 2019, up 6.7 percent from 2018. In Bangladesh there are 402.56 million livestock and poultry from which 7.51 million metric ton meat is produced in 2019 (DLS, 2019). The quality of meat (e.g. color, pH, drip loss, cooking loss, CP, EE, Ash etc.) is technologically and economically important not only for food-processing industry but also for consumers as an important attribute during purchasing meat (Akhter et al., 2009; Baset et al., 2003; Bithi et al., 2020; Disha et al., 2020; Habiba et al., 2021; Rahman et al., 2014). In Bangladesh, meat quality evaluation still depends on traditional analytical technology which involves chemical, biological and microbiological determinations but those are tedious, time-consuming, sample destructive and environmentally unfriendly (Rahman et al., 2020; Rana et al., 2014; Haque et al., 2017; Modak et al., 2009). The lack of fast, reliable and non-destructive methods for determining meat characteristics in the carcass and meat cuts has been one of the main obstacles for the development of quality control in the meat industry. Meat processing industries expect accurate, faster, real-time, low-cost and non-chemical detection technologies. In the meat industry, in order to reduce economic losses during processing, as well as to supply high-quality products consistently, quality control procedures must be carried out (Akhter et al., 2020; Liao et al., 2010; Lina et al., 2020; Sarker et al., 2021). Superior quality of these products is always demanded by consumers and is considered as a key factor for success in today's highly competitive market (Islam et al., 2019; Kamal et al., 2019; Prieto et al., 2009; Rokib et al., 2019; Saba et al., 2018). To realize these needs mentioned above and to fulfill consumers' satisfaction, it is very important to provide meat that can better meet the customers' needs and market requirements. Therefore, it is a crucial element within the meat industry to accurately assess meat and guarantee the quality and safety. Different techniques such as sensory analysis, chemical procedures and instrumental methods have been employed to provide information about meat quality (Brondum et al., 2000; Jahan et al., 2018; Islam et al., 2018). Sensory analysis is often implemented by professional staff. This method has been widely used in many food research fields. However, it is subjective, laborious, time-consuming and inconsistent. Chemical procedures and instrumental methods have been used in detecting quality attributes for a long time, which are more convenient, precise and effective than sensory analysis (Nicola et al., 2007; Siddiqua et al., 2018; Khan et al., 2017; Hashem et al., 2021). In terms of instrumental methods, pH is traditionally measured by pH meter by inserting it into the muscle directly after incision of the muscle, and colorimeters are commonly utilized for meat color evaluation. However, most of the above-mentioned techniques are destructive. In contrast to conventional methods, many novel and automatic technologies based on mechanical, optical, dielectrics, X-rays, spectroscopy, and nuclear magnetic resonance have emerged for detecting

*Corresponding Author

Professor Dr. Md. Abul Hashem
E-mail: hashem_as@bau.edu.bd

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these quality and safety attributes such as: Ultrasound technique, Spectroscopy technique, Computer vision, CT scanning etc. There are advantages and disadvantages of these methods. Ultrasound technique is rapid, non-destructive and non-polluting but it is very sensitive, easily affected by operators, measurement sites as well as the ultrasonic frequency and only detecting chemical compositions for some specific parts. Spectroscopy technique is simple, provides spectral information, and is able to detect internal attributes but limited sensitivity to minor components and complicated analysis. Computer vision providing spatial information, higher accuracy than manual inspection, able to detect external attributes and suitable for online detection but limited multi-constituent information, unable to detect internal attributes; and CT scanning is non-invasive, provides detailed images but it is expensive, requires longer evaluation time and has limited range of application. Among them, spectroscopy technique included near infrared spectroscopy (NIR), mid-infrared spectroscopy (MIR), far infrared spectroscopy (FIR) and Raman spectroscopy has been considered as one of the most promising technique (Williams, 2008). NIR spectroscopy has a great potential for the estimation of quality and safety attributes in meat and meat products in recent years, and has been considered as one of the effective and progressive techniques (Warriss, 2004). It is a potential analytical tool for sensitive and fast analysis with simplicity in sample preparation allowing a simultaneous assessment of numerous meat properties (Williams, 1987). NIRS has shown enormous potential to predict quality attributes, such as protein, fat, moisture, ash, myoglobin, pH value, water-holding capacity (WHC), color, marbling, tenderness and safety attributes (freshness, total bacterial count, adulteration) (Alomar et al., 2003). NIRS is an analytical technique that uses a source producing light of known wavelength pattern (usually 800–2500 nm) and that enables to obtain a complete picture of the organic composition of the analyzed substance/material (Kempen, 2001). It is based on the principle that different chemical bonds in organic matter absorb or emit light of different wave lengths when the sample is irradiated. Nowadays NIRS is widely and successfully used in many different fields, also for feed and food analysis. However, standard methods for quality evaluation of meat products, such as color and pH, are often rather imprecise and time consuming. Near-infrared spectroscopy (NIRS) is fast, reliable, accurate and inexpensive (Prevolnik et al., 2004; Teye et al., 2013) and has great potential as a substitute for chemical composition analysis of meat and meat products (Andres et al., 2007).

Recent developments in microfabrication and miniaturization of optical systems have allowed the creation of “palm-sized” spectrophotometers which are compact, mobile and can be carried in a pocket. To our knowledge, only one study has been conducted by Dixit et al (2020) by mini NIR spectroscopy for predicting intramuscular fat in lamb. In the current study, a “palm-sized” mini NIR spectrophotometer has been used for predicting physicochemical traits of beef combined with Chemometrics. Therefore, the objective of the present study was to evaluate the feasibility of mini NIRS to predict drip loss, cooking loss, color and proximate components in beef.

Materials and Methods

Experimental site

This experiment was conducted at Meat Science Laboratory under the Department of Animal Science, Bangladesh Agricultural University, Mymensingh.

Meat samples preparation

In total, 60 samples of *M. longissimus thoracis* (between the 12th and 13th ribs) were collected from 60 carcasses of indigenous cattle from a local market in the Mymensingh district of Bangladesh at 7.00 am. The meat samples were immediately transferred to the Animal Science Meat Laboratory, Bangladesh Agricultural University, Mymensingh in an icebox. The collected meat samples were weighed and stored in a refrigerator for 24 hours at 4°C for further analysis. After 24 hours, the samples were removed from the refrigerator and then kept on a tray for about 10-12 minutes to release absorbed moisture and then used for NIR spectra collection.

NIR spectra acquisition

NIR reflectance spectra were collected using mini NIR (DLP NIRscan Nano EVM, Texas Instruments Inc., Texas, USA). Mini NIR works in the wavelength range of 900-1700 nm with an optical resolution of 10 nm. A scan configuration specifies the following parameters of a scan: Wavelength range: Start and End wavelengths (in nm) or spectral range of interest for the scan. The minimum wavelength is 900 nm and the maximum wavelength is 1700 nm. Width in nm: This number must be greater than 8 nm and corresponds to the desired smallest wavelength content that needed to resolve in a scan. The DLP NIRscan Nano optical resolution is 10 nm, so values less than 10 nm result in lower signal intensity. Number of patterns: This number defines how many wavelength points are captured across the defined spectral range. Depending on the previous setting, the GUI computes the maximum number of patterns and indicates them as the "Max Limit." Number of scans to average: This is the repeated back-to-back scans that are averaged together. Refrigerated beef samples were thawed and subjected to spectroscopic analysis while packed in ziplock plastic bags. For NIR analysis, the sample window of mini NIR was placed over the meat samples (60) surface and the spectra were collected at 3 different locations. Each spectrum was recorded at room temperature in the NIR region of 900 to 1700 nm with a spectral increment of about 3.90 nm between the contiguous bands, thus producing a total of 228 bands.

Chemical and physical attributes analyses

Proximate components such as dry matter (DM), Ether Extract (EE), crude protein (CP) and ash were carried out according to the methods (AOAC, 1995). All analyses were done in triplicate and the mean value was reported. The differences in weight between the fresh and dried samples represent the water content. An oven (GALLENKAMP Hot Box Oven with Fan - Size 2 CHF097 XX2.5) was used for determining dry matter kept for 24 hrs at 105°C. Crude protein (CP) was determined using total nitrogen content of each sample by using kjeldahl apparatus. Ether extract content was determined by Soxhlet apparatus using diethyl ether. Total ash content was measured using muffle furnace where porcelain crucibles with samples were heated at 550°C for 6 hours, then cooled inside desiccators and weighed. Cooking loss was measured by using a hot water bath and food-grade thermometer at 71°C internal temperature of meat sample. Cooking loss was determined following the methods of Haque et al. (2017) and Rima et al. (2019). Drip loss was determined following the methods of Arain et al. (2010).

The pH was collected on intact *M. longissimus thoracis* using a Hanna PH meter (Hanna Instruments HI99163 Portable HACCP Compliant pH Meter for Meat); three replicates were taken on each sample and averaged to form the final pH reading.

Color was measured on *M. longissimus thoracis* after 24 h of freezing at 4° C using a Konica Minolta Chroma Meters CR-410; 3 consecutive readings were recorded on meat samples and averaged to form the final color reading for each sample. Meat color was expressed according to the CIE-Lab color space by reporting lightness (L*), redness (a*), and yellowness (b*) (CIE, 1978).

Saturation index (SI) was calculated as $SI = \sqrt{a^{*2} + b^{*2}}$ and hue angle (H) as $H = \tan^{-1}(b^*/a^*)$ (AMSA, 1991).

Model development and spectral data analysis

Calibrations and predictions of beef samples based on full spectra (228 variables) were established using partial least-squares regression (PLSR). The calibration models were strictly built using the calibration dataset and validated with cross-validation technique using The UNSCRAMBLER program (version 9.7.0, Camo, Trondheim, Norway). In this study, leave-one-out (i.e. full) cross-validation method was employed to validate the PLSR models. This was done by removing one sample (test sample) from the data set and PLSR model was then established for the remaining samples (training sample). Finally, the model was used to predict the sample left out. This procedure was repeated for every sample in the data set, giving a more realistic measure of the predictive errors of the model. The precision and the predictive capabilities of the models were evaluated by the coefficients of determination (R^2), root-mean-square error of calibration (RMSEC) and the root-mean-square error estimated by cross-validation (RMSECV). The best model was selected for each attribute on the basis of the highest determination coefficient (R^2) and lowest standard error of calibration (RMSEC) and validation (RMSECV). The R^2 and RMSEC or RMSECV are defined as follows:

$$R^2 = 1 - \frac{\sum_{i=1}^N (\hat{y}_i - y_i)^2}{\sum_{i=1}^N (\hat{y}_i - \bar{y}_i)^2} \quad (1)$$

$$RMSEC \text{ or } RMSECV = \sqrt{\frac{\sum_{i=1}^N (\hat{y}_i - y_i)^2}{N}} \quad (2)$$

Where \hat{y}_i = predicted value of the i^{th} sample, y_i = measured value of the i^{th} sample, N = number of samples.

Moreover, to assess the practical utility of the prediction models the ratio performance deviation (RPD) and the range error ratio (RER) were calculated. The RPD was calculated as the ratio of the standard deviation (SD) to the SECV of a given trait and the RER was calculated as the ratio between the range and the SECV of the trait (Edney et al., 1994). Ratio performance deviation values higher than 10 are considered equivalent to reference method (Williams and Sobering, 1993) and values above 2.5 are considered adequate for analytical purposes (Sinnave et al., 1994). The RER is a method of standardizing the SECV by relating it to the range of the reference data. For example, RER values of less than 6 indicate very poor classification and are not recommended for any application; RER values between 7 and 20 classify the model as poor to fair and indicate the model could be used for screening purposes, and RER values between 21 and 30 indicate a good classification suggesting the model would be suitable for a role in quality control systems ((Williams and Norris, 2001).

Results and Discussion

Reference values for physicochemical attributes of longissimus dorsi muscle of beef

Compositional values of beef samples are given in Table 1, expressed on a fresh basis, that is, in the same form as samples were scanned. To provide an overview of the structure of the samples used in the investigation, the mean, range, standard deviation (SD), coefficient of variance (CV) for color parameters (L*, a*, b*), pH, drip loss (DL%), cooking loss (CL%), dry matter (DM%), moisture (%), crude protein (CP%), ether extract (EE%) and ash (%) for all muscles determined by laboratory reference methods are summarized in Table 1.

Table 1. Descriptive statistics of chemical composition in the tested beef muscle samples

Parameter	Mean ± SD (%)	CV (%)	Range (Min-Max)
L*	42.70 ± 3.23	7.56	37.47- 49.97
a*	14.31 ± 1.59	11.11	10.78 - 17.62
b*	11.51 ± 1.60	13.9	8.01 - 15.41
pH	5.93 ± 0.09	1.52	5.80 - 6.13
DL (%)	3.35 ± 0.49	14.62	2.46 - 4.65
CL (%)	28.44 ± 2.26	7.95	24.97 - 32.58
DM (%)	25.67 ± 2.35	9.15	21.17 - 28.60
Moisture (%)	76.87 ± 1.07	1.39	73.17 - 78.87
CP (%)	22.60 ± 1.91	8.45	17.98 - 25.62
EE (%)	2.29 ± 0.82	35.81	0.7 - 3.60
Ash (%)	1.21 ± 0.41	33.88	0.83 - 2.47
H	38.81 ± 45.17	51.36	36.61 - 41.17
SI	18.36 ± 2.25	17.79	13.43-23.4

L*=lightness, a*=redness, b*=yellowness, H= hue angle, SI=saturation, DL = drip loss, CL = cooking loss, DM = dry matter, CP = crude protein, EE = ether extract

To provide an overview of the physicochemical attributes of the samples used in the investigation, the mean, range, standard deviation, and coefficient of variation for drip loss (DL %), cooking loss (CL %), moisture (%), dry matter (DM %), crude protein (CP %), ether extract (EE %) and ash (%) for all muscles determined by laboratory reference methods are summarized in Table 1. Average moisture, dry matter, protein, ether extract and ash contents of beef from longissimus dorsi muscle were found as 76.87, 25.67, 22.60, 2.29, and 1.21 %, respectively in this study. Meat color (L*, a*, b*) was quite normal. In case of a*, mean value was 14.71, CV was 11.11 and highest value was 17.62 and lowest value was 10.78. Maximum, minimum and mean value was 15.41, 8.01 and 11.51 respectively in case of b*. The L* values averaged 42.70 and ranged from 37.47-49.97 which is considered normal for beef (Chambaz et al., 2001). Maximum value of pH was 6.13 and minimum was 5.8. The average pH value was 5.93. The range is considered normal for beef (Tarrant and Mothersill, 1977).

For cooking loss mean value was 28.44, SD 2.26, CV 7.95, the maximum value was 24.97 and the minimum value was 32.58. According to Marchi et al. (2013), the values were 37.65, 3.71, 9.85, 28.58 - 45.69 for mean, SD, CV, and range respectively, regarding cooking loss.

The coefficient of variation of all measured attributes suggesting good variability of the data except pH (1.52%) and moisture (1.39%). The low variability of pH has been confirmed by several authors in beef (Andres et al., 2008), chicken (De Marchi et al., 2007), and pork (Liao et al., 2010).

Development of calibration model based on NIR spectra

NIRS calibration and prediction statistics for color parameters (L*, a*, b*), pH, drip loss (%), cooking loss (%), dry matter (%), moisture (%), crude protein (%), ether extract (%) and ash (%) of cattle longissimus dorsi muscle are presented in Table 2. Spectral data at full wavelength range (900-1700 nm) with 228 variables were modeled using linear multivariate method of PLSR. The performance of the calibration models was optimized by leave-one-out cross-validation. The detailed results of PLSR are listed in Table 2, where for PLS model, R²_c, R²_{cv}, R_p, RMSEC, and RMSECV reported. In this study, the calibration results were highly correlated to cross-validation results. The similarity in model performance implied that the models did not over-fit data, and the majority of the variance presented in the measured values was reproduced in the prediction model. Based on model performance in terms of R²_c, R²_{cv}, RMSEC and RMSECV, it seems that the PLSR model was reasonably appropriate for predicting the physicochemical attributes of beef.

Table 2. NIRS calibration and prediction statistics for different parameters

Attributes	Calibration		Cross-validation		SD	RPD	RER
	R ² _c	RMSE _c	R ² _{cv}	RMSE _{cv}			
L*	0.985182	0.389605	0.964377	0.604081	3.23	5.35	20.69
a*	0.985254	0.190950	0.964164	0.297676	1.59	5.34	22.97
b*	0.986923	0.181840	0.970539	0.272936	1.60	5.87	27.11
pH	0.985507	0.010392	0.959176	0.017441	0.09	5.16	18.92
DL (%)	0.981477	0.065773	0.952277	0.105575	0.49	4.64	20.74
CL (%)	0.982551	0.296645	0.956259	0.469669	2.26	4.81	16.20
Moisture	0.968167	0.190024	0.910547	0.318539	1.07	3.36	17.89
Dry Matter	0.978851	0.339258	0.954681	0.496621	2.35	4.73	14.96
Crude Protein	0.982020	0.253490	0.948502	0.429006	1.91	4.45	17.80
Ether Extract	0.981292	0.110906	0.958342	0.165494	0.82	4.95	17.52
Ash	0.983211	0.052103	0.947915	0.091772	0.41	4.47	17.87

R²_c = calibration coefficient; R²_{cv} = cross-validation coefficient; SD = standard deviation; RMSE_c = standard error of calibration; RMSE_{cv} = standard error of cross-validation; RPD = residual standard deviation; RER = range error ratio.

Predictive ability of the PLS model is assessed by co-efficient of determination of cross-validation (R²_{cv}) and root mean square error of cross-validation (RMSE_{cv}). The best model for each trait is selected on the basis of the highest co-efficient of determination of cross-validation (R²_{cv}) and the lowest root mean square error of cross-validation (RMSE_{cv}). R²_{cv} 0.96, 0.96, 0.95, 0.96, 0.95, 0.96, 0.95, 0.91, 0.94, 0.95 and 0.94 for L*, a*, b*, pH, drip loss, cooking loss, DM, moisture, CP, EE and ash respectively; and RMSE_{cv} values are 0.60, 0.29, 0.28, 0.02, 0.11, 0.47, 0.50, 0.32, 0.43, 0.17 and 0.09 for L*, a*, b*, pH, drip loss, cooking loss, DM, moisture, CP, EE and ash respectively. So results above values indicate that PLS model has prediction ability. R²_c for moisture and DM were 0.96, 0.98 which considered normal (Tøgersen et al., 1999; Alomar et al., 2003)

To assess the practical utility of the prediction models the ratio performance deviation (RPD) and the range error ratio (RER) were calculated. RPD were 5.35, 5.34, 5.87, 5.16, 4.64, 4.81, 4.73, 3.36, 4.45, 4.95 and 4.47 for L*, a*, b*, pH, drip loss, cooking loss, DM, moisture, crude CP, EE and ash respectively. It indicates that all values are adequate for analytical purposes as values above 2.5 are adequate for analytical purposes (Sinnave et al., 1994). Range error ratio were 20.69, 22.97, 27.11, 18.92, 20.74, 16.20, 14.96, 17.89, 17.80, 17.52 and 17.87 for L*, a*, b*, pH, drip loss, cooking loss, dry matter, moisture, crude protein, ether extract and ash respectively. The range of the reference data, RER value between 7-20 classify the model as poor to fair and indicate it could be used for screening purposes and RER values between 21 and 30 indicate a good classification suggesting the model could be suitable for application in quality control (Williams and Norris, 2001). So for the above RER values the model is fair to good.

To visualize graphically the performance of the PLS calibration models, the measured value and its predicted values resulting from the optimal PLS models are plotted and displayed in Figures 1, 2, 3, 4, 5 and 6.

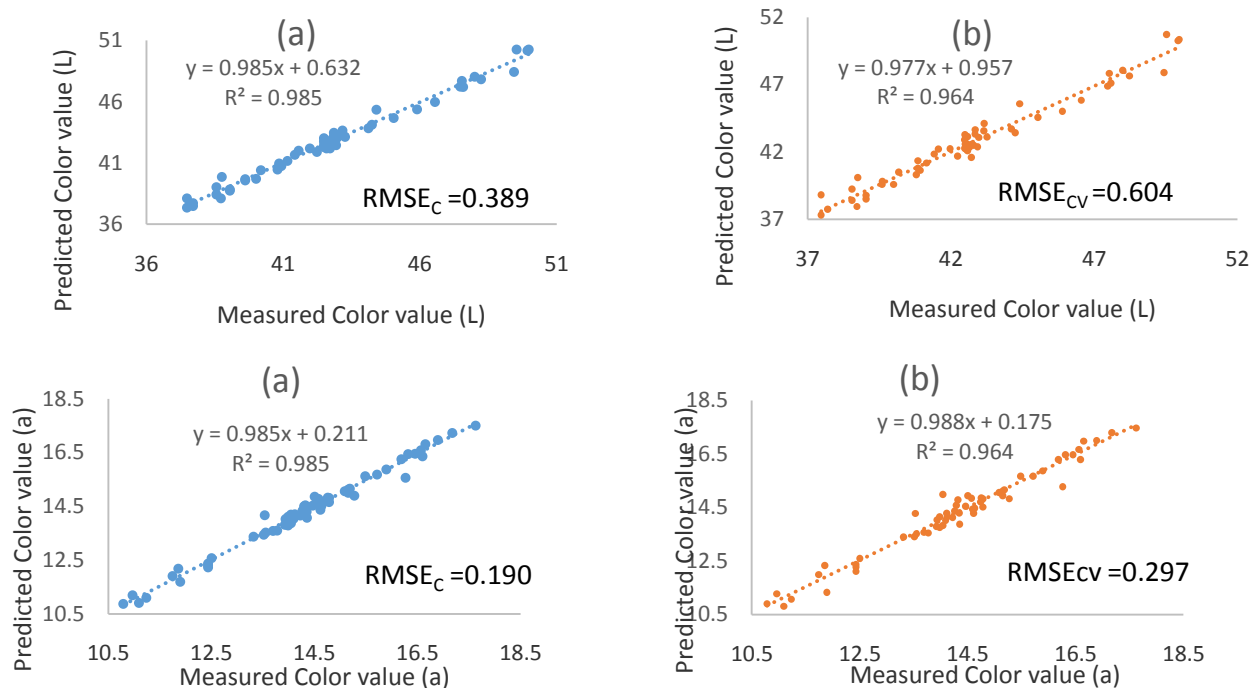


Figure 1. Prediction of L* and a* using PLS model for (a) calibration and (b) cross validation.

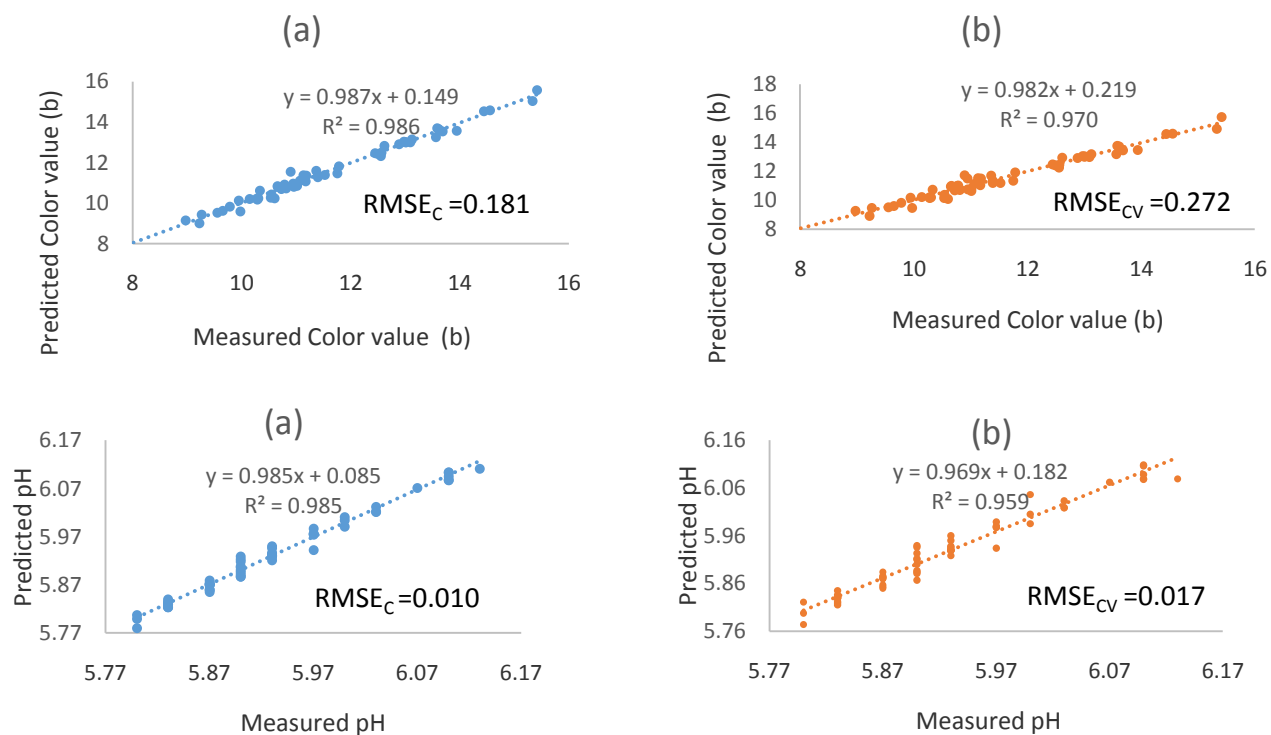


Figure 2. Prediction of b* and pH using PLS model for (a) calibration and (b) cross validation.

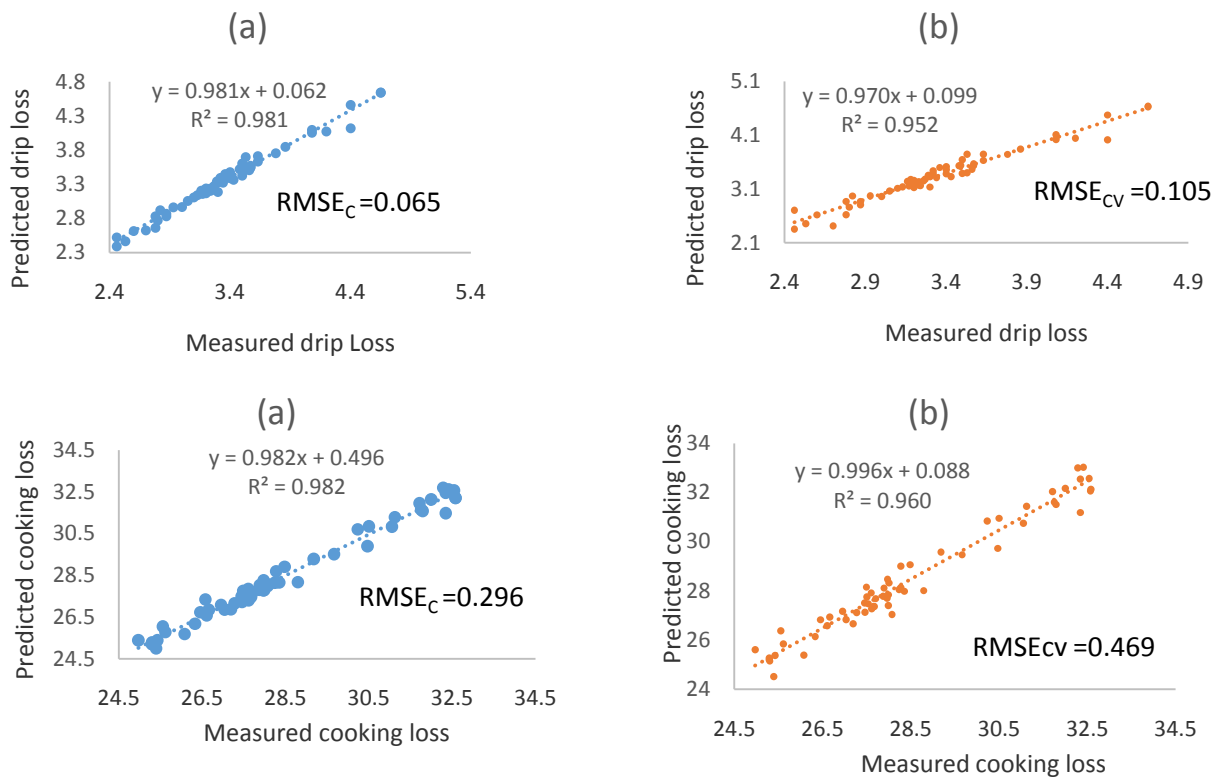


Figure 3. Prediction of drip and cooking loss using PLS model for (a) calibration and (b) cross validation.

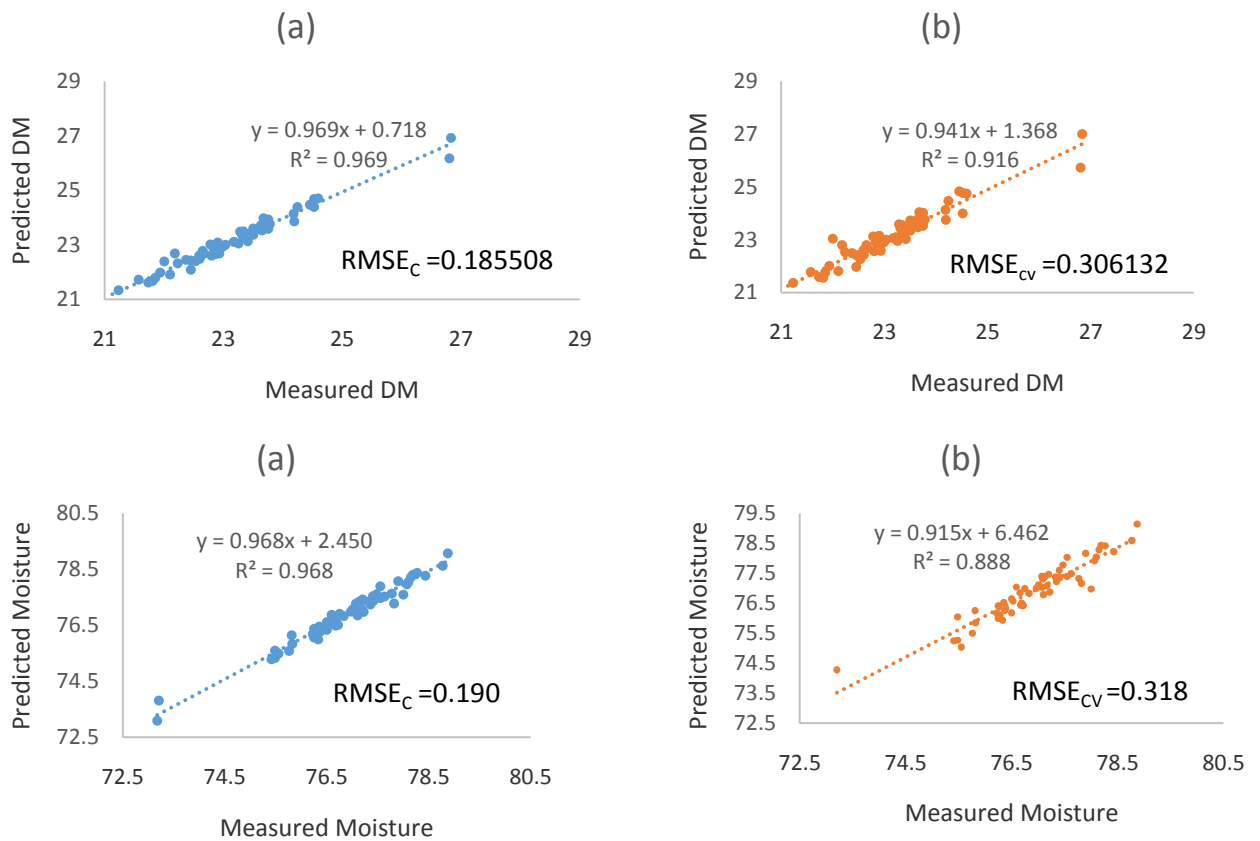


Figure 4. Prediction of DM and moisture using PLS model for (a) calibration and (b) cross validation.

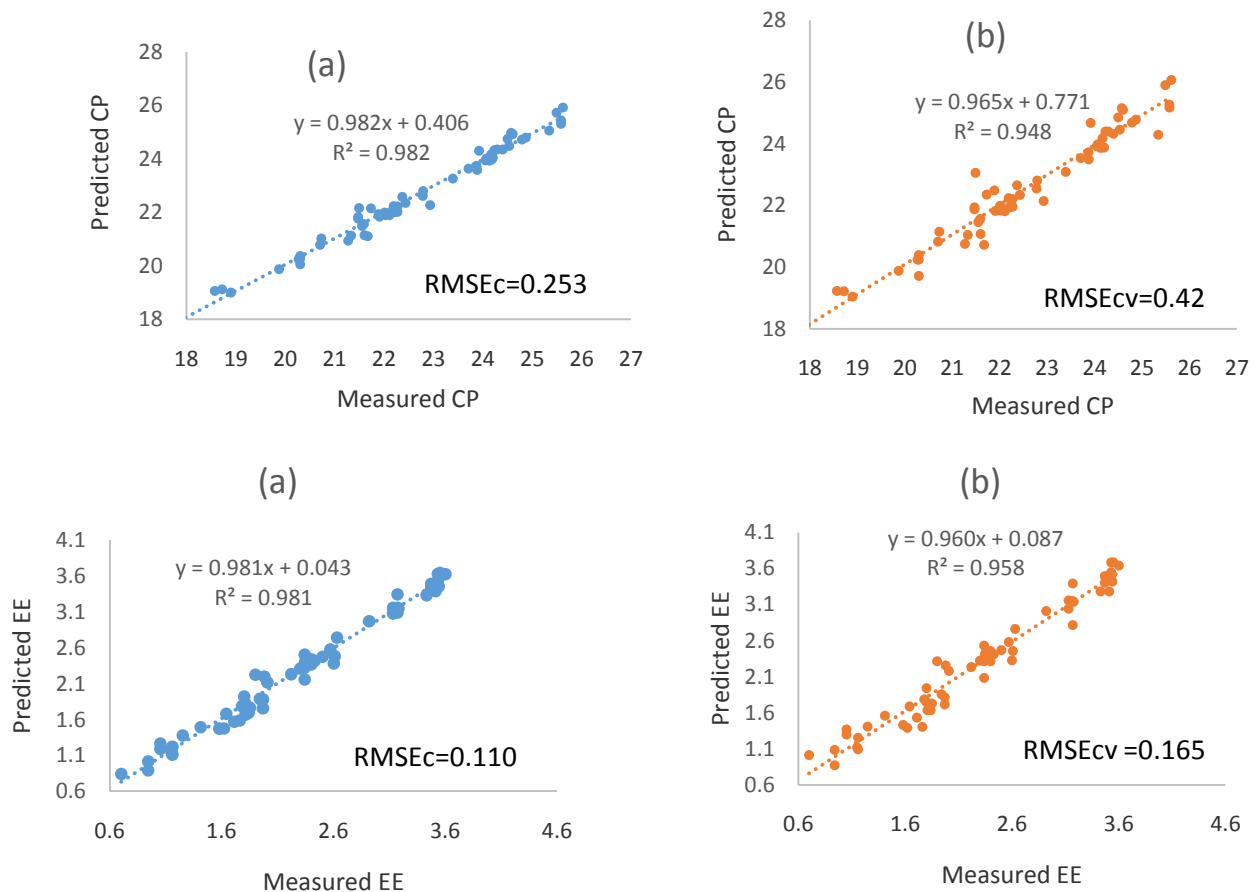


Figure 5. Prediction of CP and EE using PLS model for (a) calibration and (b) cross validation.

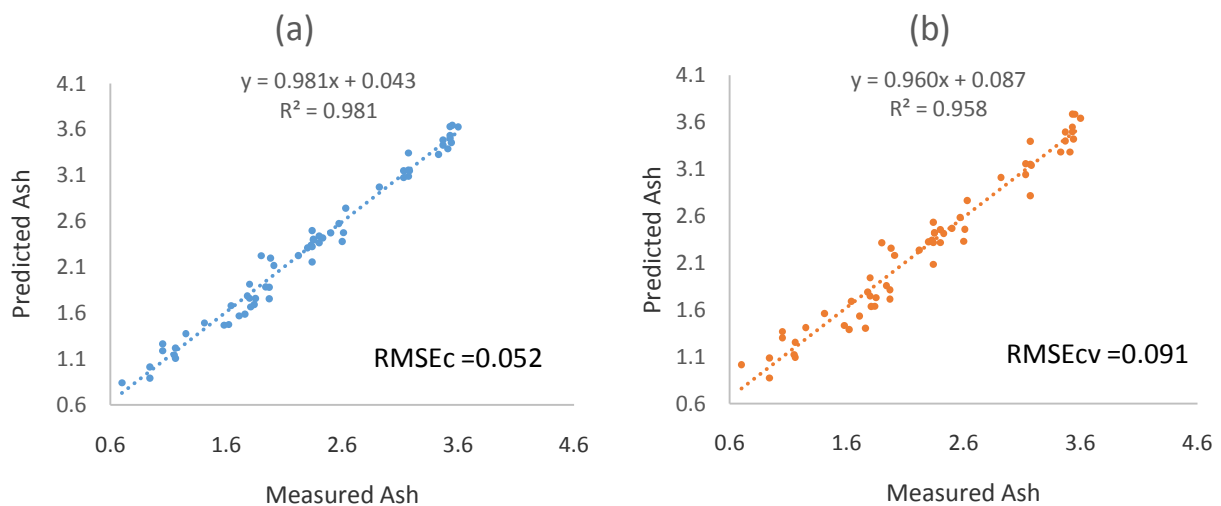


Figure 6. Prediction of Ash using PLS model for (a) calibration and (b) cross validation.

The above graphs show the values of regression models. The higher the value of R^2 (near to 1.00) indicates the higher the accuracy of the models. From the established PLSR models, we obtained coefficients of determination (R^2_{cv}) of 0.96, 0.96, 0.97, 0.95, 0.95, 0.95, 0.91, 0.91, 0.94, 0.95, and 0.91 L*, a*, b*, pH, drip loss, cooking loss, dry matter, moisture, CP, EE, and ash respectively. Generally, the accuracy (i.e. the closeness between actual and the predicted values) of regression model is considered as excellent when the $R^2 \geq 0.90$ (Kamruzzaman et al., 2015). Therefore, the developed model can be considered sufficiently accurate for future application.

Conclusions

This study was conducted to investigate the ability of mini near infrared (NIR) spectroscopy for prediction of beef quality traits. This study revealed that NIR spectroscopy coupled with PLSR can be successfully utilized as a rapid screening technique to predict the beef quality. From the established PLSR models, we obtained coefficients of determination (R^2_{cv}) value more than

0.90 for all all measurable traits. The results showed that mini NIR spectroscopy has potential as a non-destructive and rapid method to predict beef quality attributes.

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Declaration of Conflicting Interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Ethical Approval

This article does not contain any studies with human participants or animals performed by any of the authors. Meat samples were collected from the slaughterhouse.

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