RESEARCH REPORT

pork checkoff

PORK QUALITY

Title: Post-harvest Prediction of Pork Tenderness – **NPB #09-243**

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Date Submitted: 6/1/2011

Industry Summary:

To ensure high eating quality of pork, the industry has relied on "enhancing" the product by pumping it with a solution of phosphates, salt and sometimes flavorings. This approach has been relatively successful, however, there are growing indications that the demand for non-enhanced pork products is increasing and would be considered a premium product. Thus, the ability to select pork that is "naturally" tender for use in a non-enhanced product line would increase the marketing opportunities for pork and the profitability of pork production. A system for noninvasive prediction of pork loin tenderness has been developed and tested under some laboratory and commercial conditions. The present experiments were conducted to provide a broad-scale test of the efficacy of this system across multiple packing plants representing a diversity of carcass chilling methods, including both conventional spray chilling and blast chilling. Twelve hundred eight boneless pork loins were evaluated on the loin boning and trimming line of four large-scale commercial plants and tenderness was predicted with a noninvasive technology. Vacuum-packaged loins were aged and tenderness was measured objectively with slice shear force at 15 days after harvest, which is approximately the average time between harvest and retail consumption of loin chops. The prediction system allowed for identification of a group of loins that were more consistently tender. The system was efficacious both among and within packing plants. This technology could both facilitate tenderness-based pork merchandising systems and serve as a research tool to facilitate improvement of pork tenderness. Consequently, use this technology should help the industry to improve consumer satisfaction and demand for pork.

Key Words: Pork, Tenderness, Prediction, Instrument, Near-Infrared, Slice Shear Force

Scientific Abstract:

The present experiments were conducted to provide a broad-scale test of the efficacy of a non-invasive system for pork loin tenderness prediction across multiple packing plants representing a diversity of carcass chilling methods. Exposed LM on the ventral side of boneless loins was evaluated, with visible and near-infrared spectroscopy (VISNIR), at line speed on the loin boning and trimming lines of large-scale commercial plants (Exp. 1, n = 1,208, 4 plants; Exp. 2, n = 599, 3 plants). Boneless loin sections were aged (2°C) and fresh (never frozen) chops were cooked (71°C) and LM slice shear force (SSF) was measured at 15 d postmortem. In Exp.

1, on-line classification of pork loins, based on spectroscopic evaluation and prediction of SSF with a previously developed model, resulted in VISNIR tenderness classes that differed in mean LM SSF values at 15 d postmortem ($P < 10^{-11}$). Relative to loins predicted to be tender, loins that were not predicted to be tender were more likely to have SSF > 25 kg ($P < 10^{-9}$). While these findings were favorable, further examination of the data showed that the prediction model only accounted for approximately one-half of the variation in plant means for SSF (r = 0.73). To develop a robust model that would properly reflect tenderness variation among and within packing plants, correlation analysis was conducted for each plant to identify the wavelength range at which reflectance was most highly related to SSF. For each plant, the strongest correlation was found at or near 822 nm. Also, variation in the plant means for reflectance at 822 nm accounted for virtually all of the variation in plant means for SSF (r = -0.99). That is reflectance at 822 nm was indicative of variation in tenderness both among and within plants. Predicted tenderness classes, based on reflectance at 822 nm, differed in mean LM SSF values at 15 d postmortem ($P < 10^{-26}$) and the percentage of loins with SSF > 25 kg ($P < 10^{-16}$). Exp. 2 confirmed that use of this VISNIR system with reflectance at 822 nm would provide a robust method to classify pork loins for tenderness, as VISNIR predicted tenderness classes differed greatly in mean LM SSF values at 15 d postmortem ($P < 10^{-9}$) and the percentage of loins with SSF > 25 kg ($P < 10^{-5}$). These results clearly indicate that the VISNIR technology could be used to non-invasively classify pork loins on-line for tenderness.

Introduction:

Historically, pork longissimus has been considered to be relatively tender (DeVol et al., 1988. However, available data contradict the assumption that pork is uniformly tender. It has been reported that there was significant animal-to-animal variation in pork tenderness (Davis et al., 1975; DeVol et al., 1988). Furthermore, selection for increased lean growth in pork has been associated with unfavorable changes in the rate of postmortem pH decline, postrigor calpastatin activity, and tenderness (Lonergan et al., 2001). Moeller et al. (2009) reported that there was a strong relationship between Warner-Bratzler shear force and consumer like ratings of nonenhanced and enhanced pork loins, suggesting that there was substantial variation in tenderness of U.S. pork loins and that tenderness was the major driver of consumer satisfaction. To ensure high eating quality of pork, the industry has relied on “enhancing” the product by pumping it with a solution of phosphates, salt and sometimes flavorings. This approach has been relatively successful (Moeller et al.; 2009), however, there are growing indications that the demand for non-enhanced pork products is increasing and would be considered a premium product. Thus, the ability to select pork that is “naturally” tender for use in a non-enhanced product line would increase the marketing opportunities for pork and the profitability of pork production. A system for noninvasive prediction of pork loin tenderness has been developed and tested under some laboratory and commercial conditions (Shackelford et al., 2011b). The present experiments were conducted to provide a broad-scale test of the efficacy of this system across multiple packing plants representing a diversity of carcass chilling methods, including both conventional spray chilling and blast chilling.

Objectives:

The present experiments were conducted to provide a broad-scale test of the efficacy of this system across multiple packing plants representing a diversity of carcass chilling methods, including both conventional spray chilling and blast chilling.

Materials & Methods.

Loins sampled in this experiment were obtained from federally-inspected packing plants and did not involve animals originating from or under the control of the U.S. Meat Animal Research Center (USMARC). Therefore, animal procedures were not reviewed and approved by the USMARC Animal Care and Use Committee..

Experiment 1. Boneless pork loins (n = 1,208) were evaluated on the loin boning and trimming line of four large-scale commercial plants (n = 300 or 304/plant). Exposed LM on the ventral side of boneless loins was evaluated with VISNIR using a commercial system that was developed for on-line evaluation of beef tenderness. The spectroscopy systems were described by Shackelford et al. (2011d) and LM slice shear force (**SSF**) was predicted in real time using the model of Shackelford et al. (2011b). Although not possible for one plant, an attempt was made to evaluate each loin with two instruments (Table 1).

Boneless loins were vacuum-packaged, boxed, and transported (-2.8ºC) to USMARC. All loins arrived at USMARC within 6 to 12 hours of boning and were immediately refrigerated (1.5ºC), unboxed, weighed for subsequent determination of purge loss, and aged. At 14 days postmortem, loins were unpackaged allowed to drip for 5 minutes and weighed for determination of purge loss. Two 2.54-cm thick chops were obtained from the 11th rib region of each loin. The following day (i.e., 15 days postmortem), fresh (never frozen) chops were cooked (71°C) with a belt grill and LM slice shear force (**SSF**) was measured on each of the two chops (Shackelford et al., 2004). The duplicate SSF values were averaged and that value was used for all analyses.

Experiment 2. To evaluate models developed in Experiment 1, boneless pork loins (n = 599) were evaluated on the loin boning and trimming line of three large-scale commercial plants on two sampling days. Loins were processed in a similar manner to Experiment 1 and details of loin selection and processing are reported elsewhere (Shackelford et al., 2011a). Because loin selection took place in multiple plants at the same time, for two of the three plants, only a single instrument was tested (Table 1).

Statistical analysis

Analysis of variance for mean SSF differences among data sets (Figure 1) or plants (Figure 2) was conducted using the GLMMIX procedure of SAS and differences among least-squares means were determined with the diff and lines options. Variances were compared among data sets (Figure 1) or plants (Figure 2) using the F-Test Two-Sample for Variances analysis tool of Microsoft Excel 2007. Analysis of variance for mean SSF differences among VISNIR tenderness classes (Figures 3 and 4 and Table 2) was conducted using the Single Factor ANOVA tool of Microsoft Excel 2007. Differences in the frequency of loins with SSF values > 25 kg were compared with chi-square analysis and Bonferroni correction for multiple comparisons using the Categorical data option of the Compare2 program of WinPepi (Version 11.4; www.brixtonhealth.com/pepi4windows.html). Correlation analysis was conducted with the Corr procedure of SAS.

Results and Discussion:

The present data sets (Exp. 1 and 2) contained ample variation in SSF at 15 d postmortem for system validation (Figure 1). The mean SSF, the level of variation in SSF, and the percentage of samples with SSF > 25 kg were greater ($P < 0.05$) than was observed in development of the system to predict pork LM SSF with VISNIR (Shackelford et al., 2011b). There was a high degree of difference among plants in mean SSF, the level of variation in SSF, and the percentage of samples with SSF > 25 kg ($P < 0.05$; Figure 2). Shackelford et al, (2011a) objectively compared tenderness differences among three plants and ascribed the variation in tenderness among plants to differences in chilling rate. Specifically, blast chilling resulted in a higher frequency of loins with excessively high (> 25 kg) LM SSF values. This is particularly relevant to the present discussion because the data set that was used to develop the VISNIR technology for pork LM SSF prediction did not include any carcasses from plants with blast chilling. Nonetheless, in Exp. 1, on-line classification of pork loins based on spectroscopic evaluation of the ventral side of LM and prediction of SSF with the model of Shackelford et al. (2011b) resulted in VISNIR tenderness classes that differed in mean LM SSF values at 15 d postmortem ($P < 10^{-11}$; Figure 3). Relative to loins predicted to be tender by spectroscopic evaluation of the ventral side of LM, loins that were not predicted to be tender were more likely to have SSF > 25 kg ($P < 10^{-9}$). While these findings were favorable, further examination of the data showed that the prediction model did not properly account for tenderness differences among plants. Variation in the plant means for predicted SSF accounted for approximately one-half of the variation in plant means for SSF (r = 0.73). Thus, a new model was developed from the present data set.

Because the VISNIR system measures reflectance across a wide range of wavelengths, it can detect very subtle differences among data sets which might not have been directly related to the trait of interest. For instance, because of differences among plants in the layout of the loin boning and trimming lines, the time interval between deboning and application of VISNIR was not the same in each plant. A difference between plants of as little as 2 min in the time interval (i.e., 3 min vs 5 min) between deboning and application of VISNIR will result in a large enough difference in the proportion of myogbin in the oxymyoglobin state that the spectra will differ significantly among plants. Given the very large tenderness differences among plants and the confounded differences in bloom time before VISNIR, development of a multifactor partial least squares regression model would likely have resulted in an over specified model. To develop a robust model, correlation analysis was conducted for each plant to identify the wavelength range at which reflectance was most highly related to SSF. For each plant, the strongest correlation was found at or near 822 nm. For plants 1, 2, and 3, lower reflectance at 822 nm was associated with higher SSF ($P < 0.01$; Table 2) and the degree of SSF difference among predicted tenderness classes was greater when loins were sorted based on reflectance at 822 nm than using the model of Shackelford et al. (2011b). Also, variation in the plant means for reflectance at 822 nm accounted for virtually all of the variation in plant means for SSF ($r = -0.99$). That is reflectance at 822 nm was indicative of variation in tenderness both among and within plants. Consequently, when samples were classified for tenderness across all samples based on reflectance at 822 nm, the resulting classes differed in mean LM SSF values at 15 d postmortem ($P < 10^{-26}$) and the percentage of loins with SSF > 25 kg ($P < 10^{-16}$; Bottom Panel of Figure 3). Comparison of the results from sorting loins for tenderness based on the model of Shackelford et al. (2011b) with the results based on reflectance at 822 nm (top and bottom panels of Figure 3, respectively) indicates that use of reflectance at 822 nm would result in a more effective classification system.

To confirm that use of this VISNIR system with reflectance at 822 nm would provide a robust method to classify pork loins for tenderness, Exp. 2 was conducted. As in Exp. 1, loins were classified as VISNIR predicted tender if the observed reflectance at 822 nm was greater than the median. Again, VISNIR predicted tenderness classes differed greatly in mean LM SSF values at 15 d postmortem ($P < 10^{-9}$) and the percentage of loins with SSF > 25 kg ($P < 10^{-5}$; Figure 4).

To help elucidate the basis for the VISNIR tenderness sorting, correlation coefficients were determined for the relationships of predicted slice shear force, based on reflectance at 822 nm, and observed slice shear force to meat quality traits among and within packing plants for Exp. 1 and Exp. 2 (Table 3). Across all samples in Exp 2., sarcomere length and IMF were negatively correlated ($P < 0.001$) with both predicted SSF and observed SSF. For sarcomere length, the negative correlation was significant ($P < 0.01$) for loins sampled from each plant. Results for IMF were inconsistent across plants. For some traits, the direction of the correlation was the opposite for predicted SSF as compared to observed SSF. For example, ultimate pH was positively correlated ($P < 0.001$) with VISNIR predicted SSF in both Exp. 1 and 2; however, ultimate pH was negatively correlated ($P < 0.05$) with observed SSF in both Exp. 1 and 2. Cooking loss was negatively correlated ($P < 0.001$) with VISNIR predicted SSF in both Exp. 1 and 2; but, cooking loss was positively correlated ($P < 0.001$) with observed SSF in both Exp. 1 and 2. In summary, these correlations show a moderate relationship between VISNIR tenderness sorting, based on reflectance at 822 nm, and traits related to ultimate pH/lean color/water-holding capacity but the relationship does not provide a causative explanation for the mechanism of VISNIR tenderness sorting. However, sarcomere length data are consistent with the observation that beef carcasses predicted to be tender by VISNIR have longer sarcomeres in LM, gluteus medius, semimbranosus, and biceps femoris (Shackelford et al., 2011cd).

Consistent with the relationship between VISNIR predicted SSF and purge loss (Table 3), results of models developed to predict purge loss from VISNIR were highly correlated with VISNIR predicted SSF (data not shown in tabular form). Consequentially, it was determined that there was not any added benefit of implementing models for prediction of purge loss.

Shackelford et al. (2011b) reported that 62% of the variation in LM IMF could be accounted for by spectroscopic evaluation of the ventral side of LM. That model accounted for 45% of the variation in LM IMF in Exp. 2 (Figure 5). While the lower r^2 obtained in Exp. 2 suggested that the model did not perform as well as observed previously, comparison of the residual SD suggested that the model performed slightly better in Exp. 2. Whereas Shackelford et al. (2011b) observed a residual SD of 0.63%, the residual SD was only 0.57% in Exp. 2. The observed bias between predicted and observed LM IMF was only 0.16% suggesting that both procedures for VISNIR application and IMF determination were robust over time.

The present experiment resulted in development of a method to noninvasively predict LM tenderness of pork loins that was efficacious both among and within packing plants. This technology could both facilitate tenderness-based pork merchandising systems and serve as a research tool to facilitate improvement of pork tenderness.

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Table 1. Number of sampling d on which loins were evaluated by visible and near infrared (VISNIR) spectroscopy and number of loins sampled stratified by processing plant and sampling day.

Experiment	d	Plant	Instruments	Number of loins sampled
1	1	Plant 1	5015	300
1	2	Plant 2	5015 and 5016	300
1	3	Plant 3	5015 and 5016	304
1	4	Plant 4	5015 and 5016	304
				Total = 1,208
2	5	Plant 1	5015	100
2	5	Plant 2	5016	99
2	5	Plant 5	5015 and 5016	100
2	6	Plant 1	5015	100
2	6	Plant 2	5016	100
2	6	Plant 5	5015 and 5016	100
				Total = 599

[a] 5015 and 5016 are two identical spectroscopy systems, which were custom made for tenderness prediction (Shackelford et al., 2011).

Table 2. Slice shear force (kg) as affected by sorting of pork loins, for each plant and pooled across plants, into two predicted tenderness classes based on either the model of Shackelford et al. (2011b) or relectance at 822 nm.

Plant	Predicted tender	Not predicted tender	SEM	P < F
Tenderness classes based on either the model of Shackelford et al. (2011b)				
Plant 1	13.13 (n = 150)	14.14 (n = 150)	0.30	< 0.05
Plant 2	18.77 (n = 150)	18.92 (n = 150)	0.51	ns
Plant 3	19.74 (n = 152)	21.61 (n = 152)	0.50	< 0.01
Plant 4	13.99 (n = 152)	14.71 (n = 152)	0.31	< 0.1
All plants pooled	15.71 (n = 604)	18.05 (n = 604)	0.24	$< 10^{-11}$
Tenderness classes based on relectance at 822 nm				
Plant 1	12.92 (n = 150)	14.36 (n = 150)	0.29	< 0.001
Plant 2	17.89 (n = 150)	19.79 (n = 150)	0.50	< 0.01
Plant 3	19.14 (n = 152)	22.21 (n = 152)	0.50	$< 10^{-4}$
Plant 4	14.07 (n = 152)	14.64 (n = 152)	0.31	ns
All plants pooled	15.09 (n = 604)	18.67 (n = 604)	0.23	$< 10^{-26}$

Table 3. Correlation coefficients for the relationships of predicted slice shear force, based on reflectance at 822 nm, and observed slice shear force to meat quality traits among and within packing plants for Exp. 1 and Exp. 2.

	Experiment 1									
	Correlation with predicted slice shear force					Correlation with observed slice shear force				
	All plants	Plant 1	Plant 2	Plant 3	Plant 4	All plants	Plant 1	Plant 2	Plant 3	Plant 4
Purge loss, %	-0.22***	-0.39***	-0.11	-0.09	-0.42***	0.18***	0.06	0.37***	0.24***	0.16**
ultimate pH	0.36***	0.56***	0.23***	0.23***	0.61***	-0.09**	0.04	-0.16**	-0.12*	0.00
L*	-0.49***	-0.59***	-0.34***	-0.37***	-0.56***	-0.16***	-0.02	0.13*	-0.18**	-0.01
a*	0.01	0.05	-0.02	-0.02	-0.02	-0.04	0.00	-0.16**	0.02	0.04
b*	-0.10***	-0.37***	-0.13*	-0.08	-0.22***	-0.01	-0.09	-0.09	-0.07	-0.03
Lean color score[1]	0.21***	0.31***	0.25***	0.24***	0.20***	0.03	-0.05	-0.08	0.10	-0.01
Marbling score[2]	-0.13***	-0.06	-0.22***	-0.28***	0.02	-0.16***	-0.14*	-0.20***	-0.21***	-0.01
Cooking loss, %	-0.36***	-0.59***	-0.26***	-0.16**	-0.63***	0.29***	0.16**	0.53***	0.55***	0.09

	Experiment 2							
	Correlation with predicted slice shear force				Correlation with observed slice shear force			
	All plants	Plant A	Plant B	Plant C	All plants	Plant A	Plant B	Plant C
Purge loss, %	-0.34***	-0.50***	-0.06	-0.42***	0.24***	0.06	0.44***	0.01
ultimate pH	0.47***	0.61***	0.12	0.59***	-0.10*	-0.01	-0.36***	0.21**
L*	-0.49***	-0.53***	-0.19**	-0.58***	-0.02	-0.10	0.26***	-0.33***
a*	-0.17***	-0.10	-0.20**	-0.21**	-0.02	0.13	-0.13	-0.07
b*	-0.06	-0.04	0.02	-0.17*	-0.04	-0.12	0.04	0.02
Lean color score[1]	0.29***	0.32***	0.10	0.34***	0.05	0.15*	-0.15*	0.26***
Marbling score[2]	-0.04	0.11	-0.40***	-0.03	-0.13**	0.02	-0.32***	-0.04
Intramuscular fat, %	-0.18***	-0.01	-0.48***	-0.21**	-0.22***	-0.08	-0.43***	-0.15*
Cooking loss, %	-0.40***	-0.54***	0.08	-0.50***	0.24***	0.16*	0.59***	-0.04
Sarcomere length, µm	-0.29***	-0.36***	-0.29**	-0.28**	-0.60***	-0.65***	-0.57***	-0.62***
Desmin, % degraded	-0.06	-0.15	-0.33***	0.09	-0.42***	-0.39***	-0.42***	-0.37***

[1] 1 =pale, pinkish gray; 6 = dark purplish-red

[2] 1 = devoid; 10 = abundant

$^{*}P < 0.05$

$^{**}P < 0.01$

$^{***}P < 0.001$

Figure 1. Frequency distributions of LM slice shear force at 15 d postmortem for Exp. 1 and 2 as compared to the frequency distributions of LM slice shear force at 14 d postmortem for the data set (Shackelford et al., 2011b) that was used to develop the VISNIR model for prediction of SSF with visible and near infrared (VISNIR) spectroscopic evaluation of either the ventral side of LM.

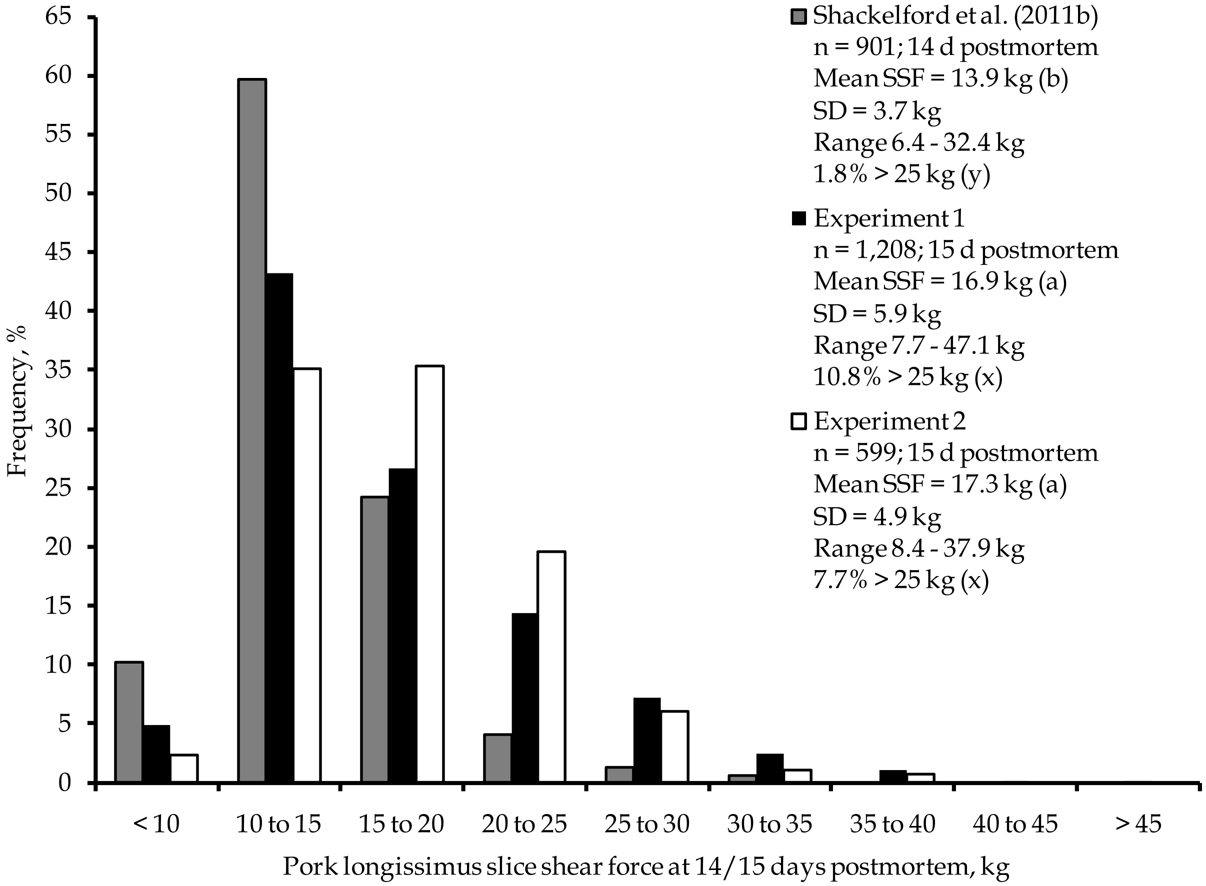
Figure 2. Frequency distributions of LM slice shear force at 15 d postmortem for the four plants sampled in Exp. 1.

Figure 3. Exp. 1. Prediction of pork LM slice shear force using visible and near infrared (VISNIR) spectroscopic evaluation of either the ventral side of LM and the model of Shackelford et al. (2011b, top panel) or reflectance at 822 nm (bottom panel).

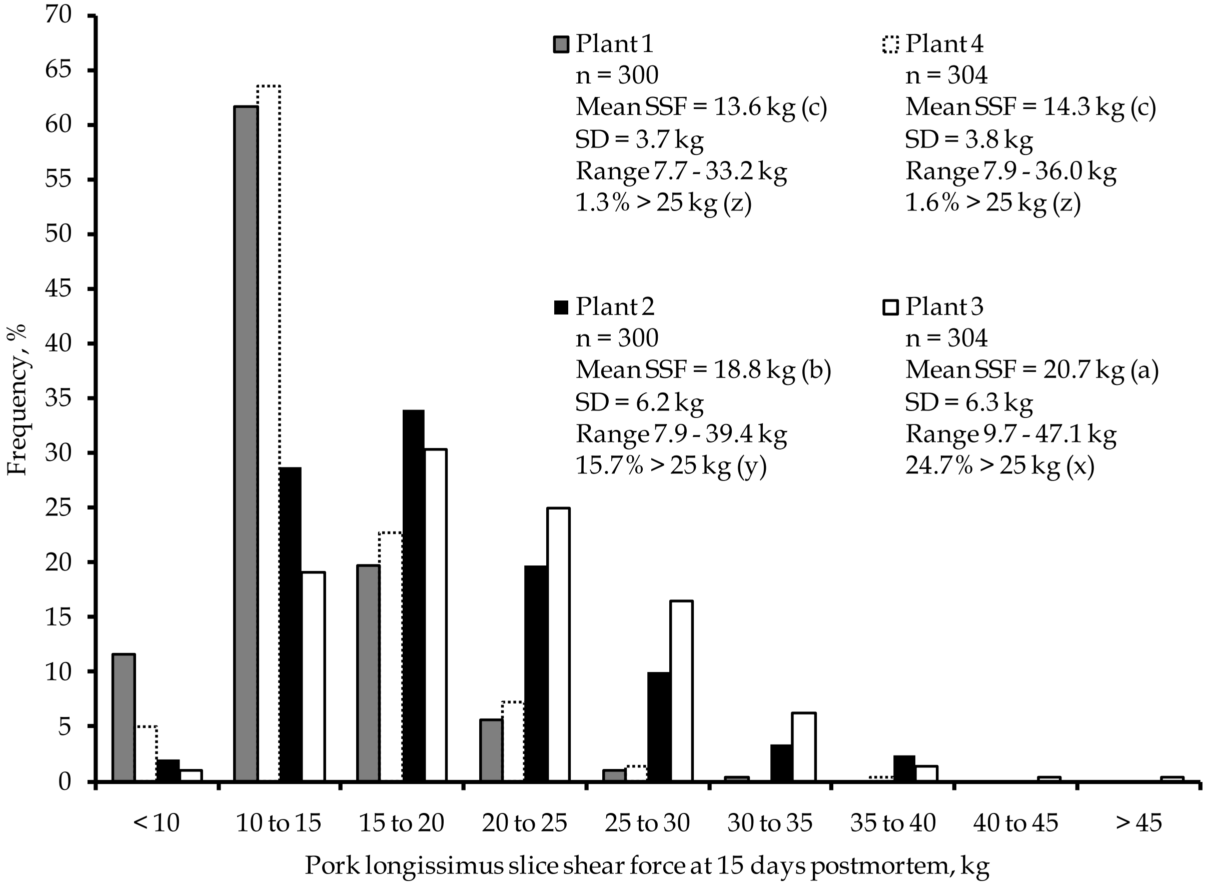
Figure 4. Exp. 2. Validation of the relationship between reflectance at 822 nm, from visible and near infrared (VISNIR) spectroscopic evaluation of the ventral side of LM, and pork LM slice shear force.

Figure 5. Exp. 2. Validation of the model of Shackelford et al. (2011b) for prediction of LM intramuscular fat percentage using visible and near infrared (VISNIR) spectroscopic evaluation of the ventral side of LM. RSD = Residual standard deviation.

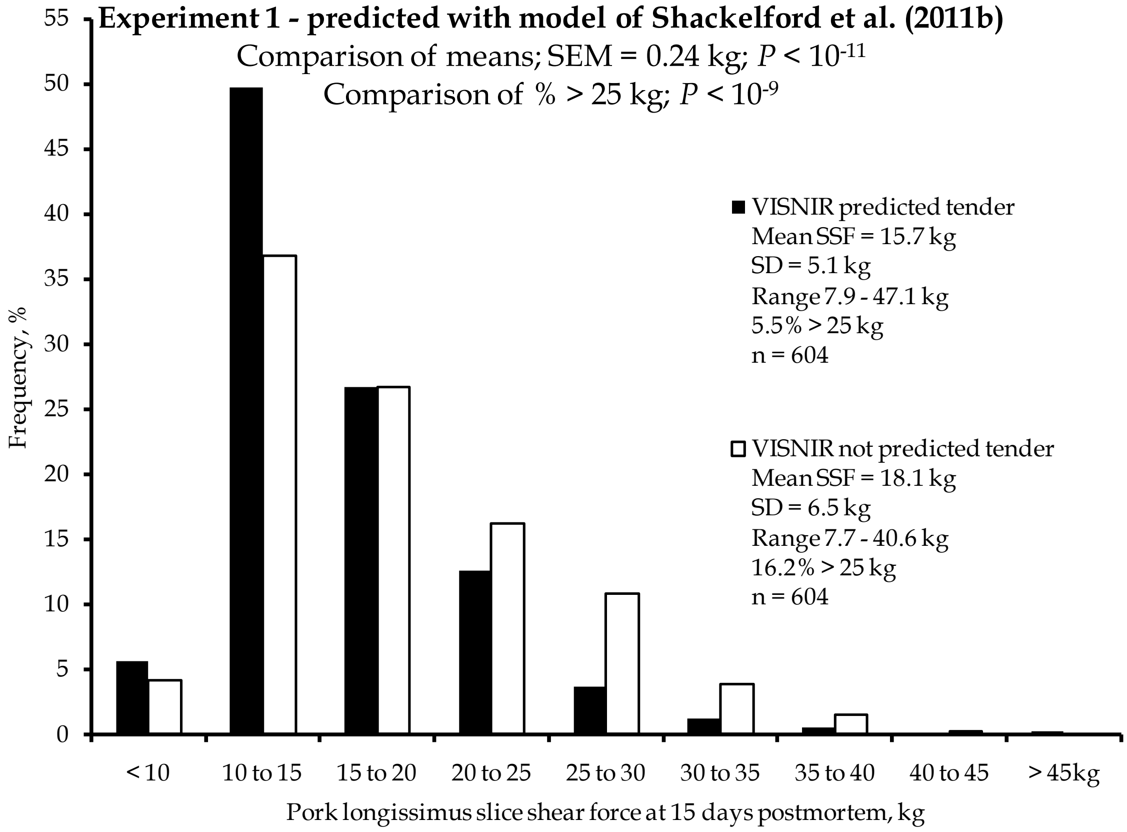
Shackelford et al. Figure 1



Shackelford et al. Figure 2



Shackelford et al. Figure 3



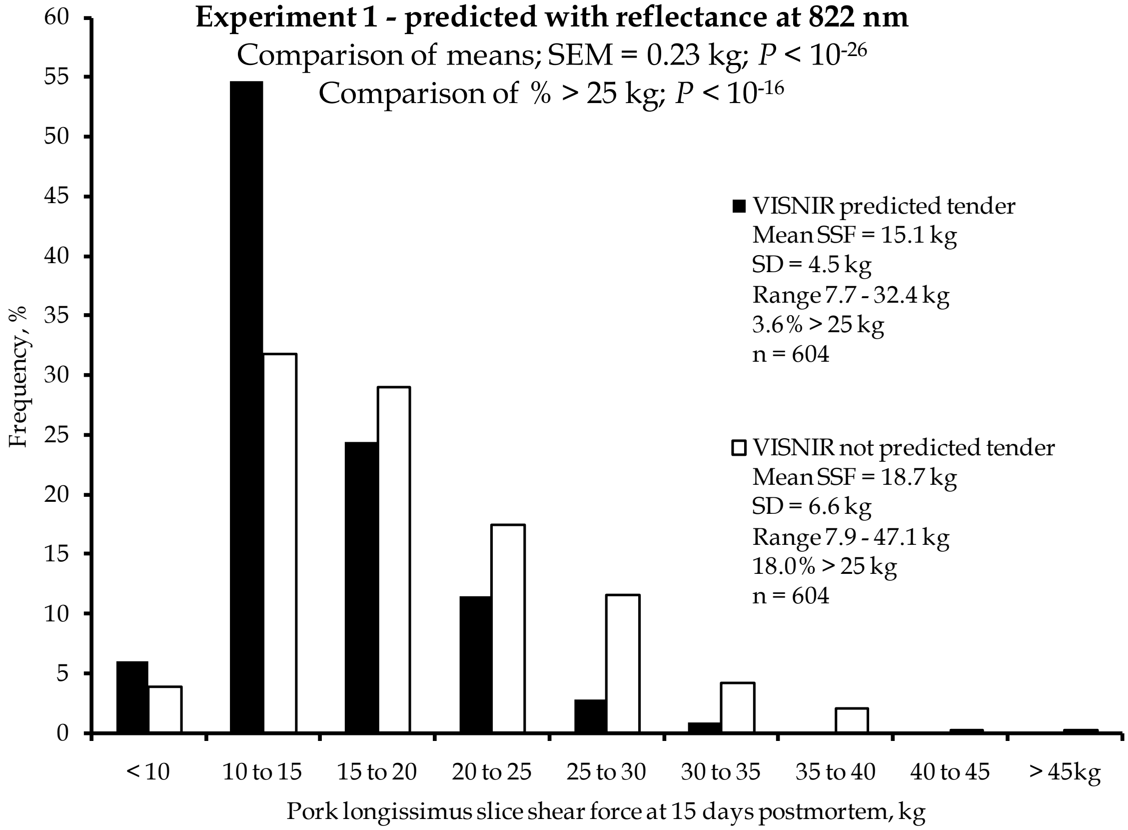
Experiment 1 - predicted with model of Shackelford et al. (2011b)

Comparison of means; SEM = 0.24 kg; $P < 10^{-11}$

Comparison of % > 25 kg; $P < 10^{-9}$

■ VISNIR predicted tender
Mean SSF = 15.7 kg
SD = 5.1 kg
Range 7.9 - 47.1 kg
5.5% > 25 kg
n = 604

□ VISNIR not predicted tender
Mean SSF = 18.1 kg
SD = 6.5 kg
Range 7.7 - 40.6 kg
16.2% > 25 kg
n = 604



Experiment 1 - predicted with reflectance at 822 nm

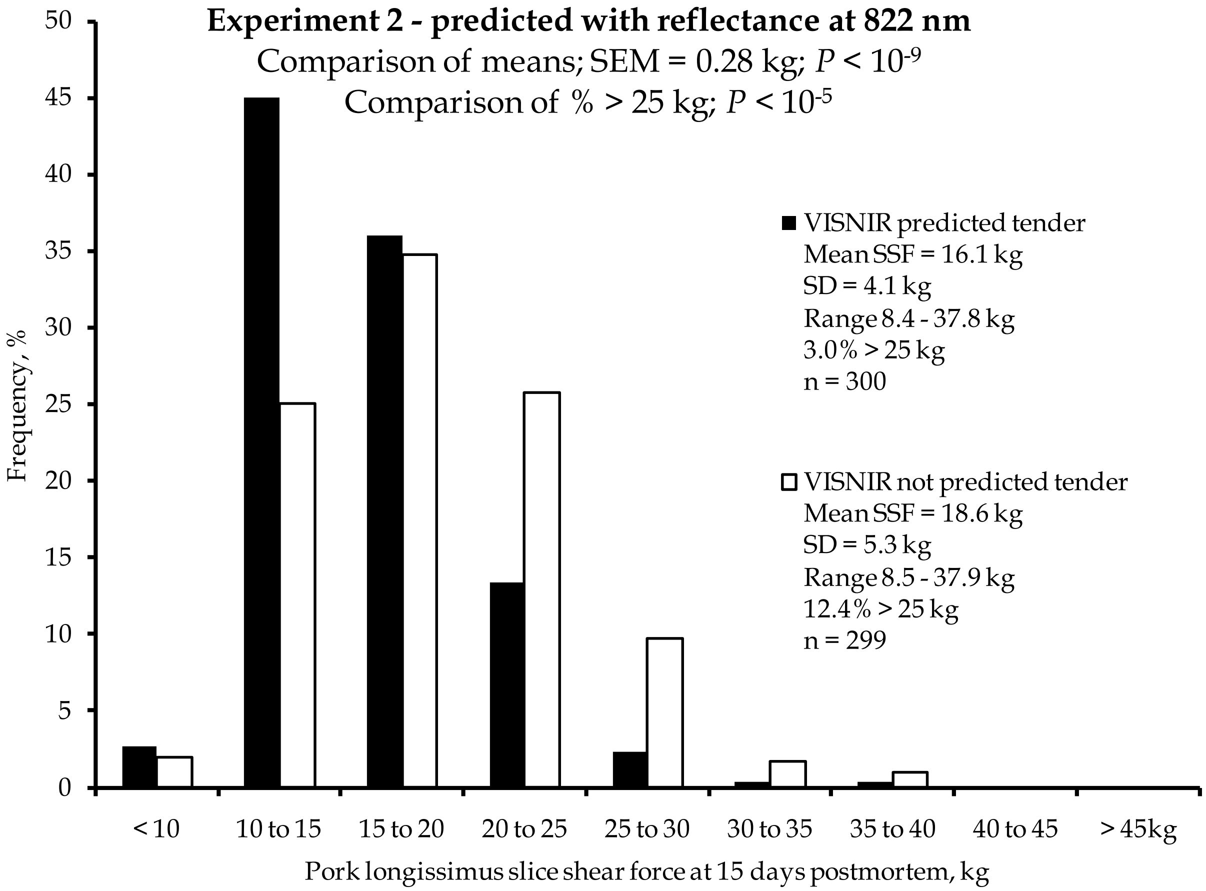
Comparison of means; SEM = 0.23 kg; $P < 10^{-26}$

Comparison of % > 25 kg; $P < 10^{-16}$

■ VISNIR predicted tender
Mean SSF = 15.1 kg
SD = 4.5 kg
Range 7.7 - 32.4 kg
3.6% > 25 kg
n = 604

□ VISNIR not predicted tender
Mean SSF = 18.7 kg
SD = 6.6 kg
Range 7.9 - 47.1 kg
18.0% > 25 kg
n = 604

Shackelford et al. Figure 4



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Shackelford et al. Figure 5