USING SERUM CHEMISTRY PROFILES TO PREDICT BEEF TENDERNESS FOR THE PURPOSE OF ON-LINE INSTRUMENT GRADING

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Introduction

United States Department of Agriculture (USDA) quality grades are designed to sort and assign beef carcasses into grades of expected eating quality, using evaluations of carcass maturity and marbling. The vast majority (97%) of carcasses within the fed steer and heifer population are classified into the "A-maturity" group (McKenna et al., 2002). Furthermore, McKenna et al., (2002) reported that 77% of all carcasses from the fed steer and heifer population had marbling scores of "small" or "slight". With the majority of the fed steer and heifer slaughter in a narrow maturity range and in a narrow marbling range, the USDA quality grades do not effectively segregate these carcasses into uniform palatability groups. Recent research by Shanks (2002) examined 24 constituents (minerals, enzymes, and hormones) in beef blood at the time of exsanguination and found that meat tenderness could be predicted using a combination of several of these blood components. However, the Shanks (2002) study utilized only 20 animals. To validate this technique, it needed to be tested on a large number of randomly-selected animals, similar to that which would be found under normal grading conditions in a typical beef packing plant.

Objective

To determine if serum chemistry profiles are useful for predicting beef tenderness for the purpose of on-line instrument grading.

Methodology

Data were obtained from 286 head of cattle on five different dates from cattle harvested at Cargill Meat Solutions facilities. Three collections occurred at the Schuyler, NE plant and two collections occurred at the Plainview, TX plant. Cattle were selected randomly at 5-min intervals on the production line.

Serum Analysis

Blood samples were collected immediately following exsanguination, chilled in an ice bath for 2-4 hours, and centrifuged (3400 rpm) for 15 min to separate serum. Serum

was then immediately transported to the South Dakota State University Animal Disease Research and Diagnostic Laboratory and analyzed using a COBAS MIRA (F. Hoffmann-La Roche Ltd., Basel, Switzerland) spectrophotometer chemistry analyzer for nineteen compounds (Table 1). Frozen serum samples were shipped on dry ice to LINCO Research, Inc. (St. Charles, MO) to be assayed for glucagon and cortisol. Cortisol was measured using a double antibody cortisol 125I-radioimmunoassay, and glucagon was measured utilizing a 125I-labeled glucagon and a glucagon antiserum to determine the level of glucagon by the double antibody/PEG technique.

Carcass Data

After a 24 hour chill, experienced evaluators determined USDA yield and quality grades. After a 90 min bloom time, muscle color was measured on the exposed longissimus at the 12th/13th rib interface using a HunterLabs MiniScan XE colorimeter (Hunter Associates Laboratory Inc., Reston, VA) with a D65 illuminant.

Slice Shear Force Determination

One 2.5-cm-thick steak was removed from the 13th rib location from each side of each carcass. Steaks were vacuum packaged individually and shipped to Cargill Meat Solutions Laboratory (Wichita, KS) where one steak from each carcass was aged for 2 days and frozen and the other steak for 14 days and frozen. Steaks were cooked on an impingement oven to an internal temperature of 70C. A 5.1-cm x 2.5-cm slice was removed from each steak and slice shear force was determined using a slice shear force apparatus.

Statistical Analysis

Data were analyzed using the GLM procedure of SAS to calculate least squares means for data from tough versus tender carcasses. A threshold value of 20 kg for 14-d slice shear force (SSF14) was used to determine "tender" versus "tough". This 20-kg threshold was lower than the 27-kg threshold reported by Wheeler et al. (2004), but was chosen because there were very few carcasses (n = 9 / 286 = 3.1%) in our data set with SSF14 greater than 27 kg. Because this research was exploratory in nature, and not intended to finalize prediction equations for use in the field, a threshold which resulted in a substantial number of "tough" carcasses was deemed critical for a valid statistical analysis. Statistical models to classify carcasses as "tender" or "tough" were developed and tested using logistic regression (Khattree and Naik, 2000). The LOGISTIC procedure of SAS was used to test various models using the SELECTION=SCORE option. Using a response variable equal to 0 for a "tender" carcass and equal to 1 for a "tough" carcass, carcasses were classified as "tough" if the estimated probability of classifying an observation as "tough" was greater than 0.112 (32/286 because we had 32 "tough" carcasses out of 286 total carcasses; Seber, 1984).

Results & Discussion

Tender carcasses (SSF14 < 20kg) had a higher (P < 0.05) proportion of steer carcasses, higher (P < 0.05) hot carcass weights, USDA Quality Grades, and a* values and lower (P < 0.05) dark cutting discount (DARK), pH and 2-d slice shear force (SSF2) values than tough carcasses (Table 2). Tender carcasses also had lower aspartate aminotransferase (AAT), calcium, non-esterified fatty acids (NEFA), phosphorus, potassium (K), and sodium (NA) levels and higher (P < 0.05) cortisol (CORT) levels than tough carcasses (Table 3).

Logistic regression models were developed to sort carcasses into either a "certified tender" or "tough" groups with a threshold value of 20 kg for SSF14 (Table 4). No sorting resulted in a 100% certification of carcasses with an 11.2% occurrence of tough steaks in "Certified Tender" carcasses. The first type of model used USDA Quality Grade as the sole variable, and resulted in 61.5% certification of carcasses with an 8.0% occurrence of tough steaks in "Certified Tender" carcasses. The second type of model used USDA Quality Grade factors as variables, with the model that resulted in the lowest percentage tough in "Certified Tender" carcasses utilizing DARK, lean maturity score (LMAT), and marbling score, which resulted in 68.2% certification of carcasses with a 7.7% occurrence of tough steaks in "Certified Tender" carcasses. The third type of model used all carcass traits as variables with the model resulting in the lowest percentage of tough steaks in "Certified Tender" carcasses utilizing a*, DARK, and LMAT, which resulted in 69.0% certification of carcasses.

The fourth type of model used serum constituent values for variables. In our opinion, the model that best combined the lowest error in "Certified Tender" and the most ease of application was the model that utilized CORT, NEFA, and K, which resulted in 65.4% certification of carcasses with an occurrence of tough steaks in 5.3% of "Certified Tender" carcasses. The fifth type of model made use of carcass traits and serum constituent values for variables. In our opinion, the model that best combined the lowest error in "Certified Tender" and the most ease of application was the model that utilized a*, DARK and LMAT, which resulted in 69.0% certification of carcasses with a 5.1% occurrence of tough steaks in "Certified Tender" carcasses. The seventh type of model employed carcass traits, serum constituent values and SSF2 for variables and as expected had the lowest occurrence of tough steaks in "Certified Tender" carcasses and the highest certification rates.

Predicting tenderness using blood chemistry was more accurate than predicting tenderness with USDA quality grades but not as accurate as SSF2. Predicting tenderness using blood chemistry was similar in accuracy to tenderness prediction using measures of muscle color.

Shanks (2002) showed that bovine serum profiles can be used as a method to predict tenderness and had a 5% error rate for certified tender beef in a study that only used 20 head. The present study's model for serum constitutes presented a 5.3% error rate for "certified tender" beef with a certification rate of 65.4% as compared to the error rate of 8.0% and certification rate of 61.5% for the model that used USDA Quality Grades. The 5.3% error rate for the serum constitutes model compares favorably against the error rates of 6.0 and 4.9% (slice shear force), 7.0 and 6.5% (colorimeter), and 12.1 and 10.3%

(BeefCam) at 70% and 60% certification level, respectively, found by Wheeler et al., (2002).

Conclusions

Predicting tenderness using blood chemistry was more accurate than predicting tenderness with USDA quality grades but not as accurate as 2-day slice shear force. Predicting tenderness using blood chemistry was similar in accuracy to tenderness prediction using measures of muscle color. Muscle color measurement could probably be applied at a much lower cost than blood chemistry, while blood chemistry has the advantage of application on live animals.

Acknowledgements

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Tables and Figures

Albumin	ALB
Alkaline Phosphatase	AP
Amylase	AMY
Aspartate Aminotransferase	AAT
Beta-hydroxy Butyric Acid	BHBA
Calcium	CA
Chloride	CL
Cortisol	CORT
Creatinine	CREA
Creatine Phosphokinase	СРК
Gamma-Glutamyltransferase	GGLT
Globulin	GLOB
Glucose	GLCS
Glucagon	GLGN
Magnesium	MG
Non-Esterified Fatty Acids	NEFA
Phosphorus	Р
Potassium	Κ
Sodium	NA
Total Bilirubin	BILI
Total Protein	PROT
Adjusted Fat Thickness	FAT
Hot Carcass Weight	HCW
Ribeye Area	REA
Kidney, Pelvic and Heart Fat	KPH
Marbling Score	MARB
Skeletal Maturity Score	SMAT
Lean Maturity Score	LMAT
Overall Maturity Score	OMAT
Dark Cutting Discount	DARK
L*	L
a*	А
b*	В
Gender	SEX
pH	PH
2-d Slice Shear Force	SSF2
14-d Slice Shear Force	SSF14

Trait	Tender (n=254)	Tough (n=32)	P-value
SEX^{a}^	0.29 ± 0.03	0.50 ± 0.08	0.0165
FAT, cm	1.30 ± 0.03	1.27 ± 0.12	0.7374
REA, cm^{2}^	89.93 ± 0.77	91.10 ± 2.13	0.6142
HCW, kg	365.19 ± 2.64	344.24 ± 7.43	0.0083
KPH, %	2.39 ± 0.03	2.36 ± 0.09	0.7283
USDA Yield Grade	2.87 ± 0.07	2.59 ± 0.18	0.1559
MARB^{b}^	409.49 ± 4.81	381.56 ± 13.56	0.0533
SMAT^{c}^	152.81 ± 1.14	154.38 ± 3.22	0.6607
LMAT^{c}^	153.58 ± 0.89	158.75 ± 2.52	0.0540
OMAT^{c}^	153.27 ± 0.93	155.31 ± 2.62	0.4626
USDA Quality Grade ⁴	686.02 ± 2.94	660.75 ± 8.28	0.0043
DARK, %	0.00 ± 0.01	0.13 ± 0.02	< 0.0001
L	39.70 ± 0.20	38.82 ± 0.55	0.1369
А	24.18 ± 0.13	21.88 ± 0.36	< 0.0001
В	20.53 ± 0.18	18.62 ± 0.50	0.0004
PH	5.40 ± 0.01	5.55 ± 0.02	< 0.0001
2-d Cooking Shrink, %	16.18 ± 0.21	17.63 ± 0.74	0.0607
SSF2	21.14 ± 0.49	30.62 ± 1.70	< 0.0001
14-d Cooking Shrink, %	15.50 ± 0.14	15.69 ± 0.39	0.6495
SSF14	14.25 ± 0.19	25.28 ± 0.54	< 0.0001

Table 2. Least squares means of carcass traits for Tender (SSF14 < 20kg) and Tough groups

$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Table 5. Least squ	ares means of carcasses tra	its for Tender (SSF14 < 20 kg	g) and Tough groups
AP, U/L 144.74 ± 3.42 148.53 ± 9.64 0.7112 AMY, U/L 18.95 ± 0.44 19.59 ± 1.23 0.6239 AAT, U/L 84.24 ± 1.58 94.97 ± 4.43 0.0232 BHBA, mg/dL 0.26 ± 0.01 0.28 ± 0.04 0.6274 CA, mg/dL 9.51 ± 0.09 10.09 ± 0.26 0.0390 CL, mmol/L 103.56 ± 0.49 106.26 ± 1.41 0.0723 CORT, µg/dL 6.43 ± 0.13 5.26 ± 0.36 0.0023 CPK, U/L 593.53 ± 33.01 611.28 ± 92.82 0.8571 CREA, mg/dL 1.78 ± 0.03 1.96 ± 0.10 0.0764 GGLT, u/L 33.01 ± 0.92 37.44 ± 2.59 0.1086 GLOB, g/dL 4.24 ± 0.07 4.66 ± 0.21 0.0627 GLGN, pg/mL 683.22 ± 44.65 761.06 ± 125.80 0.5603 GLCS, mg/dL 189.23 ± 5.56 214.56 ± 15.66 0.1285 MG, mEq/L 2.10 ± 0.02 2.20 ± 0.06 0.1447 NEFA, mEq/L 0.20 ± 0.01 0.25 ± 0.02 0.0077 P, mg/dL 7.35 ± 0.10 8.02 ± 0.27 0.0216 K, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.0249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	Trait	Tender (n=254)	Tough (n=32)	P-value
AMY, U/L 18.95 ± 0.44 19.59 ± 1.23 0.6239 AAT, U/L 84.24 ± 1.58 94.97 ± 4.43 0.0232 BHBA, mg/dL 0.26 ± 0.01 0.28 ± 0.04 0.6274 CA, mg/dL 9.51 ± 0.09 10.09 ± 0.26 0.0390 CL, mmol/L 103.56 ± 0.49 106.26 ± 1.41 0.723 CORT, µg/dL 6.43 ± 0.13 5.26 ± 0.36 0.0023 CPK, U/L 593.53 ± 33.01 611.28 ± 92.82 0.8571 CREA, mg/dL 1.78 ± 0.03 1.96 ± 0.10 0.0764 GGLT, u/L 33.01 ± 0.92 37.44 ± 2.59 0.1086 GLOB, g/dL 4.24 ± 0.07 4.66 ± 0.21 0.0627 GLGN, pg/mL 683.22 ± 44.65 761.06 ± 125.80 0.5603 GLCS, mg/dL 189.23 ± 5.56 214.56 ± 15.66 0.1285 MG, mEq/L 2.10 ± 0.02 2.20 ± 0.06 0.1447 NEFA, mEq/L 0.20 ± 0.01 0.25 ± 0.02 0.0077 P, mg/dL 7.35 ± 0.10 8.02 ± 0.27 0.0216 K, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.0249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	ALB, g/dL	4.53 ± 0.04	4.64 ± 0.11	0.3281
AAT, U/L 84.24 ± 1.58 94.97 ± 4.43 0.0232 BHBA, mg/dL 0.26 ± 0.01 0.28 ± 0.04 0.6274 CA, mg/dL 9.51 ± 0.09 10.09 ± 0.26 0.0390 CL, mmol/L 103.56 ± 0.49 106.26 ± 1.41 0.0723 CORT, µg/dL 6.43 ± 0.13 5.26 ± 0.36 0.0023 CPK, U/L 593.53 ± 33.01 611.28 ± 92.82 0.8571 CREA, mg/dL 1.78 ± 0.03 1.96 ± 0.10 0.0764 GGLT, u/L 33.01 ± 0.92 37.44 ± 2.59 0.1086 GLOB, g/dL 4.24 ± 0.07 4.66 ± 0.21 0.0627 GLGN, pg/mL 683.22 ± 44.65 761.06 ± 125.80 0.5603 GLCS, mg/dL 189.23 ± 5.56 214.56 ± 15.66 0.1285 MG, mEq/L 0.20 ± 0.01 0.25 ± 0.02 0.0077 P, mg/dL 7.35 ± 0.10 8.02 ± 0.27 0.0216 K, mmol/L 7.55 ± 0.07 8.11 ± 0.20 0.0097 NA, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	AP, U/L	144.74 ± 3.42	148.53 ± 9.64	0.7112
BHBA, mg/dL 0.26 ± 0.01 0.28 ± 0.04 0.6274 CA, mg/dL 9.51 ± 0.09 10.09 ± 0.26 0.0390 CL, mmol/L 103.56 ± 0.49 106.26 ± 1.41 0.0723 CORT, µg/dL 6.43 ± 0.13 5.26 ± 0.36 0.0023 CPK, U/L 593.53 ± 33.01 611.28 ± 92.82 0.8571 CREA, mg/dL 1.78 ± 0.03 1.96 ± 0.10 0.0764 GGLT, u/L 33.01 ± 0.92 37.44 ± 2.59 0.1086 GLOB, g/dL 4.24 ± 0.07 4.66 ± 0.21 0.0627 GLGN, pg/mL 683.22 ± 44.65 761.06 ± 125.80 0.5603 GLCS, mg/dL 189.23 ± 5.56 214.56 ± 15.66 0.1285 MG, mEq/L 2.10 ± 0.02 2.20 ± 0.06 0.1447 NEFA, mEq/L 0.20 ± 0.01 0.25 ± 0.02 0.0077 P, mg/dL 7.35 ± 0.10 8.02 ± 0.27 0.0216 K, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.0249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	AMY, U/L	18.95 ± 0.44	19.59 ± 1.23	0.6239
CA, mg/dL 9.51 ± 0.09 10.09 ± 0.26 0.0390 CL, mmol/L 103.56 ± 0.49 106.26 ± 1.41 0.0723 CORT, µg/dL 6.43 ± 0.13 5.26 ± 0.36 0.0023 CPK, U/L 593.53 ± 33.01 611.28 ± 92.82 0.8571 CREA, mg/dL 1.78 ± 0.03 1.96 ± 0.10 0.0764 GGLT, u/L 33.01 ± 0.92 37.44 ± 2.59 0.1086 GLOB, g/dL 4.24 ± 0.07 4.66 ± 0.21 0.0627 GLGN, pg/mL 683.22 ± 44.65 761.06 ± 125.80 0.5603 GLCS, mg/dL 189.23 ± 5.56 214.56 ± 15.66 0.1285 MG, mEq/L 2.10 ± 0.02 2.20 ± 0.06 0.1447 NEFA, mEq/L 0.20 ± 0.01 0.25 ± 0.02 0.0077 P, mg/dL 7.35 ± 0.10 8.02 ± 0.27 0.0216 K, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.0249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	AAT, U/L	84.24 ± 1.58	94.97 ± 4.43	0.0232
CL, mmol/L 103.56 ± 0.49 106.26 ± 1.41 0.0723 CORT, µg/dL 6.43 ± 0.13 5.26 ± 0.36 0.0023 CPK, U/L 593.53 ± 33.01 611.28 ± 92.82 0.8571 CREA, mg/dL 1.78 ± 0.03 1.96 ± 0.10 0.0764 GGLT, µ/L 33.01 ± 0.92 37.44 ± 2.59 0.1086 GLOB, g/dL 4.24 ± 0.07 4.66 ± 0.21 0.0627 GLGN, pg/mL 683.22 ± 44.65 761.06 ± 125.80 0.5603 GLCS, mg/dL 189.23 ± 5.56 214.56 ± 15.66 0.1285 MG, mEq/L 2.10 ± 0.02 2.20 ± 0.06 0.1447 NEFA, mEq/L 0.20 ± 0.01 0.25 ± 0.02 0.0077 P, mg/dL 7.35 ± 0.10 8.02 ± 0.27 0.0216 K, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.0249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	BHBA, mg/dL	0.26 ± 0.01	0.28 ± 0.04	0.6274
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	CA, mg/dL	9.51 ± 0.09	10.09 ± 0.26	0.0390
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	CL, mmol/L	103.56 ± 0.49	106.26 ± 1.41	0.0723
$\begin{array}{cccccc} {\rm CREA,\ mg/dL} & 1.78 \pm 0.03 & 1.96 \pm 0.10 & 0.0764 \\ {\rm GGLT,\ u/L} & 33.01 \pm 0.92 & 37.44 \pm 2.59 & 0.1086 \\ {\rm GLOB,\ g/dL} & 4.24 \pm 0.07 & 4.66 \pm 0.21 & 0.0627 \\ {\rm GLGN,\ pg/mL} & 683.22 \pm 44.65 & 761.06 \pm 125.80 & 0.5603 \\ {\rm GLCS,\ mg/dL} & 189.23 \pm 5.56 & 214.56 \pm 15.66 & 0.1285 \\ {\rm MG,\ mEq/L} & 2.10 \pm 0.02 & 2.20 \pm 0.06 & 0.1447 \\ {\rm NEFA,\ mEq/L} & 0.20 \pm 0.01 & 0.25 \pm 0.02 & 0.0077 \\ {\rm P,\ mg/dL} & 7.35 \pm 0.10 & 8.02 \pm 0.27 & 0.0216 \\ {\rm K,\ mmol/L} & 7.55 \pm 0.07 & 8.11 \pm 0.20 & 0.0097 \\ {\rm NA,\ mmol/L} & 149.68 \pm 0.54 & 153.33 \pm 1.52 & 0.0249 \\ {\rm BILI,\ mg/dL} & 0.45 \pm 0.01 & 0.46 \pm 0.03 & 0.7168 \\ \end{array}$	CORT, µg/dL	6.43 ± 0.13	5.26 ± 0.36	0.0023
GGLT, u/L 33.01 ± 0.92 37.44 ± 2.59 0.1086 GLOB, g/dL 4.24 ± 0.07 4.66 ± 0.21 0.0627 GLGN, pg/mL 683.22 ± 44.65 761.06 ± 125.80 0.5603 GLCS, mg/dL 189.23 ± 5.56 214.56 ± 15.66 0.1285 MG, mEq/L 2.10 ± 0.02 2.20 ± 0.06 0.1447 NEFA, mEq/L 0.20 ± 0.01 0.25 ± 0.02 0.0077 P, mg/dL 7.35 ± 0.10 8.02 ± 0.27 0.0216 K, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.0249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	CPK, U/L	593.53 ± 33.01	611.28 ± 92.82	0.8571
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	CREA, mg/dL	1.78 ± 0.03	1.96 ± 0.10	0.0764
$ \begin{array}{llllllllllllllllllllllllllllllllllll$	GGLT, u/L	33.01 ± 0.92	37.44 ± 2.59	0.1086
GLCS, mg/dL 189.23 ± 5.56 214.56 ± 15.66 0.1285 MG, mEq/L 2.10 ± 0.02 2.20 ± 0.06 0.1447 NEFA, mEq/L 0.20 ± 0.01 0.25 ± 0.02 0.0077 P, mg/dL 7.35 ± 0.10 8.02 ± 0.27 0.0216 K, mmol/L 7.55 ± 0.07 8.11 ± 0.20 0.0097 NA, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.0249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	GLOB, g/dL	4.24 ± 0.07	4.66 ± 0.21	0.0627
$ \begin{array}{llllllllllllllllllllllllllllllllllll$	GLGN, pg/mL	683.22 ± 44.65	761.06 ± 125.80	0.5603
NEFA, mEq/L 0.20 ± 0.01 0.25 ± 0.02 0.0077 P, mg/dL 7.35 ± 0.10 8.02 ± 0.27 0.0216 K, mmol/L 7.55 ± 0.07 8.11 ± 0.20 0.0097 NA, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.0249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	GLCS, mg/dL	189.23 ± 5.56	214.56 ± 15.66	0.1285
P, mg/dL 7.35 ± 0.10 8.02 ± 0.27 0.0216 K, mmol/L 7.55 ± 0.07 8.11 ± 0.20 0.0097 NA, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.0249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	MG, mEq/L	2.10 ± 0.02	2.20 ± 0.06	0.1447
K, mmol/L 7.55 ± 0.07 8.11 ± 0.20 0.0097 NA, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.0249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	NEFA, mEq/L	0.20 ± 0.01	0.25 ± 0.02	0.0077
NA, mmol/L 149.68 ± 0.54 153.33 ± 1.52 0.0249 BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	P, mg/dL	7.35 ± 0.10	8.02 ± 0.27	0.0216
BILI, mg/dL 0.45 ± 0.01 0.46 ± 0.03 0.7168	K, mmol/L	7.55 ± 0.07	8.11 ± 0.20	0.0097
	NA, mmol/L	149.68 ± 0.54	153.33 ± 1.52	0.0249
PROT, g/dL 8.81 ± 0.10 9.30 ± 0.27 0.0917	BILI, mg/dL	0.45 ± 0.01	0.46 ± 0.03	0.7168
	PROT, g/dL	8.81 ± 0.10	9.30 ± 0.27	0.0917

Table 3. Least squares means of carcasses traits for Tender	(SSF14 < 20kg) and Tough groups
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	logistic regression models		D	T 1
			Percentage Tough	
Obs	# Model Variables	Certification Rate	Of those Certified	Of those Not- Certified
No Soi				
	0 UNSORTED	100.0	11.2	NA
	Quality Grade	100.0	11.2	1 1 1
	1 USDA Quality Grade	61.5	8.0	16.4
	Quality Grade Factors	01.0	0.0	10.1
	1 DARK	99.0	10.2	100.0
	2 DARK, LMAT	83.6	9.2	21.3
	3 DARK, LMAT, MARB	68.2	7.7	18.7
	4 DARK, LMAT, MARB, SMAT	67.5	7.8	18.3
	s Traits			
	1 A	66.5	5.8	22.1
	2 A, DARK	69.0	7.7	19.3
	3 A, DARK, LMAT	69.0	5.1	25.0
284	4 A, DARK, LMAT, HCW	70.1	6.5	22.4
284	5 A, DARK, LMAT, HCW MARB	73.6	5.7	26.7
284	6 A, DARK, LMAT, MARB, REA, SEX	74.3	6.2	26.0
284	7 A, DARK, LMAT, MARB, REA, SEX, B	72.5	6.8	23.1
Serum	Constituents			
286	1 CORT	59.4	6.5	18.1
286	2 CORT, NEFA	61.2	7.4	17.1
286	3 CORT, NEFA, K	65.4	5.3	22.2
285	4 CORT, NEFA, K, AAT	67.0	4.7	24.5
283	5 CORT, NEFA, K, AAT, CL	67.1	4.7	23.7
283	6 CORT, NEFA, K, AAT, CL, ALB	67.8	5.7	22.0
283	7 CORT, NEFA, K, AAT, CL, ALB, BHBA	68.2	5.7	22.2
283	8 CORT, NEFA, K, AAT, CL, ALB, BHBA, P	68.2	5.7	22.2
Carcas	s Traits and Serum Constituents			
	1 A	66.5	5.8	22.1
	2 A, DARK	69.0	7.7	19.3
	3 A, DARK, LMAT	69.0	5.1	25.0
	4 A, DARK, CORT, K	72.9	7.2	22.1
	5 A, DARK, LMAT, CORT, K	71.8	5.9	25.0
	6 A, DARK, LMAT, CORT, K, NEFA	71.8	5.9	25.0
	7 A, DARK, LMAT, CORT, NEFA, CL, BILI	72.2	5.9	24.4
-	8 A, DARK, LMAT, CORT, NEFA, CL, BILI, K	73.0	3.9	30.3
	s Traits, Serum Constituents and SSF2 Values	00.5		
	1 SSF2	90.5	5.2	31.8
	2 SSF2, DARK	90.5	4.8	36.4
	3 SSF2, DARK, CL	86.5	4.0	29.0
	4 SSF2, DARK, CL, CORT	87.0	4.5	26.7
-	5 SSF2, DARK, CL, CORT, A	84.6	3.6	28.6
	6 SSF2, DARK, CL, CORT, A, BILI	85.0	3.6	29.4
	7 SSF2, DARK, CL, CORT, A, BILI, ALB	85.0	3.1	32.4
227	8 SSF2, DARK, CL, CORT, A, BILI, ALB, SMAT	85.5	3.1	33.3

Table 4. Certification rate and percentage tough in certified and not-certified groups for various types of logistic regression models