The possibilities of in vivo predicting of intramuscular fat content in the lean cattle

J. Tomka¹, P. Polák¹, D. Peškovičová¹, E. Krupa¹, L. Bartoň², D. Bureš² ¹Animal Production Research Centre Nitra, Hlohovská 2, Lužianky, Slovak Republic ²Institute of Animal Science, Přátelství 815, Praha Uhříněves, Czech Republic

Summary

The effect of measurement location along the musculus longissimus thoracis et lumborum on in vivo intramuscular fat content prediction was evaluated in our study. Attention was also paid to effect of echocoupler usage on the prediction accuracy. There were 57 bulls (Charolais, Beef Simmental, Czech Fleckvieh, crossbreds) used in the experiment. The cross sectional scanograms from musculus longissimus thoracis et lumborum were obtained from the site between 8th and 9th rib and between 12th and 13th rib using Sonovet 2000. The animals were measured at the age of 14 to 20 months. Within computer image analysis the mean gray value of the whole muscle area (GRAY8, GRAY12) and area fraction of intramuscular fat (INT8, INT12) were calculated. The actual intramuscular fat content was assessed within laboratory analyses. Pearson's correlations between ultrasound measurements and laboratory determined intramuscular fat content were calculated. When data on whole dataset were considered, the correlations between ultrasound measurements and laboratory determined intramuscular fat content ranged from -0.03 to 0.59. When data on beef bulls were considered, the highest correlations were calculated ranging from 0.55 to 0.73. When data on crossbreds were considered the correlations between ultrasound measurements and laboratory determined intramuscular fat content ranged from -0.31 to 0.63. When data on Czech Fleckvieh bulls were considered the correlations between ultrasound measurements and laboratory determined intramuscular fat content ranged from -0.51 to 0.69. Linear regression models were designed for IMF prediction including single ultrasound measurement or the combination of two ultrasound measurements.

Key words: carcass quality; cattle; *in vivo* ultrasound method; marbling; intramuscular fat.

Introduction

The use of ultrasound is very promising method for prediction of meat characteristics. However it is very important to minimize the effects influencing the accuracy of this method. Hassen et al. (2001) stated the actual content of intramuscular fat in the muscle is one of those factors. In the other paper Hassen et al. (1999) showed that fatter the animal the higher the repeatability of measurement. Different results according to different intramuscular fat levels reported Aass et al. (2006). The correlations between ultrasound measurements and actual intramuscular fat content improved from 0.23 to 0.41 after the exclusion of bulls group with lower intramuscular fat content. Similar trend was reported in our previous work Tomka et al. (2007). We reported the correlations improved from 0.11 - 0.47 to 0.50 - 0.59 after the exclusion of the bulls group with lower intramuscular fat content. Another fact that should be considered when predicting the intramuscular fat content in live animals is the location of measurement. Albrecht et al. (2006) stated that there is different distribution of intramuscular fat along the musculus longissimus thoracis et lumborum. Faucitano et al. (2004) reported that the highest intramuscular fat content was observed in the middle of thoracis part of *musculus longissimus dorsi*. Some authors also propose connecting the echocoupler to the probe for better adjusting to round contours of bull body and to capture the whole muscle eye area.

The aim of this paper is to evaluate the effect of echocoupler usage and the location of ultrasound measurement on intramuscular fat content prediction accuracy.

Material and Methods

The study was carried out on 57 bulls (20 Czech Fleckvieh, 11 Charolais, 9 Beef Polled Simmental and 17 crossbreds). There were four datasets created according to breed and use of echocoupler (year of measurement). Animals of the first group (17 crossbreds of Piemontese, Charolais x Czech Fleckvieh) and the second group (12 Czech Fleckvieh bulls) were scanned in 2006 using echocoupler. The third group included 8 Czech Fleckvieh bulls and fourth group included 20 beef bulls (Charolais, Beef Polled Simmental). These animals were scanned in 2008 without using echocoupler. In the whole dataset (n = 57) the average age at slaughter was 495 days and average live animal weight was 602 kg. When data on crossbreds were considered the average age at slaughter was 512 days and live weight was 619 kg. When data on first group of Czech Fleckvieh bulls (n = 12) were considered the average age at slaughter was 511 days and average live weight was 599 kg. When data on the second group of Czech Fleckvieh bulls (n = 8) were considered the average age at slaughter was 480 days and average live weight was 601 kg.

The animals were scanned seven days before slaughter. The ultrasound machine SONOVET 2000 with 3.5 MHz (172 mm) linear probe was used in our experiment. In 2006 the echocoupler was attached to the probe for better adjusting to round shapes of bulls` bodies. In 2008 the echocoupler was not used in order to evaluate effect of this equipment on prediction accuracy. In both years scanograms of *musculus longissimus thoracis et lumborum* (MLTL) rib eye area were obtained from the site between 8th and 9th rib and between 12th and 13th rib. Ultrasound intensity was set to 80 – 85% as suggested in our previous work Tomka et al. 2007. The detailed dissection was performed twenty-four hours after slaughter. The meat sample was taken for laboratory determination of intramuscular fat content (value was entitled as IMF).

The ultrasound images were analysed using LUCIA software (LUCIA, 2005) for computer image analysis (CIA). The peak detection algorithm was applied as described in our previous work (Tomka et al., 2007). Also the mean gray value of measured muscle area was assessed. The values were entitled INT8, INT12 (scanogram values for percentage intramuscular fat according to the site of measurement) and GRAY8, GRAY12 (scanogram values for mean gray value of muscle area according to the site of measurement).

For statistical analysis SAS 9.1 software (SAS, 2005) was used. STAT (Basic Statistics) module and CORR (Correlation analysis) and GLM (General Linear Models) procedure were applied. Linear regression models were designed for IMF prediction. The models included one ultrasound measurement. Then the models using combination of the two ultrasound measurements at different sites (in models with highest R^2 previously) were designed.

Results and Discussion

Basic statistics for laboratory determined (LAB) and predicted (INT) intramuscular fat content and calculated gray values (GRAY) are summarized in Table 1.

| (01011) | | | | | | | | | | | |
|-----------|---------------|-------|------------|------|---------------|------|---------------|-------|-------------|-------|--|
| | Whole dataset | | Crossbreds | | Cz. Fleckvieh | | Cz. Fleckvieh | | Beef breeds | | |
| | (n=57) | | (n=17) | | (n=12) | | (n=8) | | (n=20) | | |
| | mean | SD | mean | SD | mean | SD | mean | SD | mean | SD | |
| LAB (%) | 1.87 | 0.79 | 1.36 | 0.48 | 1.91 | 0.82 | 1.99 | 0.65 | 2.23 | 0.84 | |
| INT8 (%) | 3.50 | 4.34 | 3.93 | 4.85 | 5.19 | 6.19 | 2.17 | 1.86 | 2.65 | 2.95 | |
| INT12 (%) | 2.68 | 2.79 | 1.67 | 0.89 | 1.93 | 1.20 | 4.57 | 4.04 | 3.06 | 3.36 | |
| GRAY8 | 21.30 | 23.77 | 5.11 | 2.49 | 6.76 | 4.78 | 37.55 | 16.08 | 37.29 | 28.35 | |
| GRAY12 | 24.39 | 29.92 | 9.14 | 4.61 | 11.49 | 3.95 | 50.86 | 42.69 | 31.80 | 33.59 | |
| | | | | | | | | | | | |

Table 1 Basic statistics for laboratory determined (LAB) and predicted (INT) intramuscular fat content and gray values (GRAY)

There was statistically significant difference between the average intramuscular fat content according to breed groups. The highest intramuscular fat content was observed in group of beef breeds and the lowest was observed in the group of crossbreds. When data on bulls measured using echocoupler (crossbreds and Czech Fleckvieh) were considered higher predicted values of intramuscular fat content (INT) were calculated at $8^{th} - 9^{th}$ rib site. When data on bulls measured without using echocoupler (Czech Fleckvieh and beef breeds) were considered higher predicted values of intramuscular fat content (INT) were calculated at $12^{th} - 13^{th}$ rib site. Calculated gray values were very variable and ranged from 5.11 to 37.55 (GRAY8) and from 9.14 to 50.86 (GRAY12). When data on bulls measured using echocoupler (crossbreds and Czech Fleckvieh) were considered higher gray values were calculated at $12^{th} - 13^{th}$ rib site. When data on beef breeds group were considered higher gray values were calculated at $12^{th} - 13^{th}$ rib site. When data on beef breeds group were considered higher gray values were calculated at $12^{th} - 13^{th}$ rib site. When data on beef breeds group were considered higher gray values were calculated at $12^{th} - 13^{th}$ rib site. When data on beef breeds group were considered higher gray values were calculated at $12^{th} - 13^{th}$ rib site. When data on beef breeds group were considered higher gray values were calculated at $12^{th} - 13^{th}$ rib site.

Correlations between predicted intramuscular fat content and mean gray values and laboratory determined intramuscular fat content are summarized in Table 2 and Table 3.

Table 2 Correlation coefficients between predicted (INT) intramuscular fat content, mean gray values (GRAY) and laboratory determined (LAB) intramuscular fat content (using echocoupler)

| | Crossbreds (n=17) | | | | Cz. Fleckvieh (n=12) | | | |
|--------|-------------------|-------|----------|---------|----------------------|--------|---------|--------|
| | INT12 | GRAY8 | GRAY12 | LAB | INT12 | GRAY8 | GRAY12 | LAB |
| INT8 | 0.24 | -0.10 | 0.14 | -0.31 | -0.07 | -0.31 | 0.29 | -0.09 |
| INT12 | | 0.29 | 0.84 *** | 0.50 * | | 0.79 * | 0.84 ** | 0.69 * |
| GRAY8 | | | 0.48 | 0.36 | | | 0.63 | 0.41 |
| GRAY12 | | | | 0.64 ** | | | | 0.59 |

Fisher's z-test *** P< 0.001, ** P< 0.01, * P< 0.05

When data on groups measured using echocoupler were considered low correlations were calculated between INT8 and laboratory determined intramuscular fat content (-0.31 and -0.09, respectively). Higher and statistically significant correlations were calculated between predicted IMF at site between 12th and 13th rib and actual IMF (0.50 and 0.69, respectively). Calculated correlations between gray values measured at site between 8th and 9th rib and actual IMF (0.36 and 0.41, respectively) were lower than those calculated between GRAY12 and actual IMF (0.64 and 0.59, respectively).

Different correlation coefficients according to breed were affected by actual intramuscular fat. This fact was shown by Hassen et al. (2001) and Aass et al. (2006), Aass et al. (2009). Different correlation coefficients can be also explained by breed. Reverter et al. (2000) reported different correlation coefficients in Angus group (r = 0.47 - 0.48) and Hereford group (r = 0.28 - 0.93).

When data on groups measured without using echocoupler were considered (Table 3) the higher variation of correlation coefficients was observed. Calculated correlations between predicted and actual intramuscular fat content were higher at site between 12^{th} and 13^{th} rib (0.10 and 0.65, respectively). Calculated correlations between mean gray values and actual intramuscular fat content were higher at site between 8^{th} and 9^{th} rib (0.03 and 0.73, respectively).

Table 3 Correlation coefficients between predicted (INT) intramuscular fat content, mean gray values (GRAY) and laboratory determined (LAB) intramuscular fat content (not using echocoupler)

| | Cz. Fleckvieh (n=8) | | | | Beef breeds (n=20) | | | | |
|--------|---------------------|--------|----------|-------|--------------------|----------|----------|----------|--|
| | INT12 | GRAY8 | GRAY12 | LAB | INT12 | GRAY8 | GRAY12 | LAB | |
| INT8 | 0.44 | 0.39 | 0.55 | -0.51 | 0.95 *** | 0.85 *** | 0.82 *** | 0.55 * | |
| INT12 | | 0.74 * | 0.93 *** | 0.10 | | 0.87 *** | 0.87 *** | 0.65 ** | |
| GRAY8 | | | 0.86 ** | 0.03 | | | 0.95 *** | 0.73 *** | |
| GRAY12 | | | | -0.06 | | | | 0.72 *** | |

Fisher's z-test *** P< 0.001, ** P< 0.01, * P< 0.05

Calculated correlation coefficients between ultrasound measurements and actual intramuscular fat content presented above are similar to findings of Herring et al. (1998) r = 0.20 - 0.61. Similar range of correlation values is presented by Aass et al. (2009) r = 0.26 - 0.87. Our calculated correlation coefficients between mean gray values and actual intramuscular fat content are similar to those presented by Polák et al. (2008) r = 0.00 - 0.79 and Indurain et al. (2006) r = 0.63. Brethour et al. (1994) reported lower correlation coefficients between gray values and marbling score r = 0.25.

The intercepts, regression coefficients, coefficient of determination and root mean square error of linear regression models for laboratory determined intramuscular fat content are summarized in Table 4.

| Dependent | Intercept |] | Linear regre | \mathbf{P}^2 | DMCE | | |
|---------------------|-----------|---------|--------------|----------------|-------|----------|------|
| variable | Intercept | INT12 | INT8 | GRAY12 | GRAY8 | - K | KMSE |
| | 0,87 ** | 0,26 * | - | - | - | 0,25 * | 0,41 |
| LAD | 1,48 *** | - | -0,03 | - | - | 0,10 | 0,48 |
| LAD (Crossbrads) | 0,72 ** | - | - | 0,06 | - | 0,41** | 0,37 |
| (Clossoleus) | 1,00 ** | - | - | - | 0,07 | 0,13 | 0,47 |
| | 0,67 * | - | - | 0,06 * | 0,02 | 0,42 * | 0,38 |
| LAD | 1,20 * | 0,47 * | - | - | - | 0,47 * | 0,64 |
| LAD (Crash | 1,97 *** | - | -0,01 | - | - | 0,01 | 0,85 |
| (Czech | 0,70 | - | - | 0,12 | - | 0,35 | 0,71 |
| | 1,44 ** | - | - | - | 0,07 | 0,17 | 0,78 |
| 2006) | 1,14 * | 0,04 | - | - | 0,14 | 0,71 * | 0,51 |
| LAD | 1,92 ** | 0,02 | - | - | - | 0,01 | 0,70 |
| LAD (Czash | 2,39 *** | - | -0,18 | - | - | 0,26 | 0,60 |
| Czech | 2,04 ** | - | - | -0,001 | - | 0,004 | 0,70 |
| | 1,94 * | - | - | - | 0,001 | 0,001 | 0,70 |
| 2008) | 2,27 ** | - | -0,24 | 0,004 | - | 0,33 | 0,63 |
| | 1,73 *** | 0,16 ** | - | - | - | 0,43 ** | 0,65 |
| LAB | 1,82 *** | - | 0,15 | - | - | 0,30 * | 0,72 |
| (Beef breeds) | 1,66 *** | - | - | 0,02 *** | - | 0,52 *** | 0,60 |
| | 1,43 *** | - | - | - | 0,02 | 0,53 *** | 0,59 |
| | 1,44 *** | 0,02 | - | - | 0,02 | 0,53 ** | 0,60 |

Table 4 Linear regression models for laboratory determined intramuscular fat content (LAB) using ultrasound measurements (INT12, INT8, GRAY12, GRAY8)

Fisher's z-test *** P< 0.001, ** P< 0.01, * P< 0.05,

 R^2 – coefficient of determination, RMSE – root mean square error

When data on crossbreds were considered the R^2 of linear models with single ultrasound measurement ranged from 0.10 to 0.41 and did not improve after combination of GRAY12 and GRAY8. When data on group of Czech Fleckvieh bulls measured in 2006 (with echocoupler) were considered the R^2 of linear models with single ultrasound measurement ranged from 0.01 to 0.47. The coefficient of determination of model improved after combination of INT12 and GRAY8 to 0.71. When data on group of Czech Fleckvieh bulls measured in 2008 (without echocoupler) were considered the R^2 of linear models with single ultrasound measurement varied from 0.001 to 0.26 and improved slightly after combination of INT8 and GRAY12. When data on beef breeds were considered the R^2 of linear models with single ultrasound measurement ranged from 0.30 to 0.53 and did not improved after combination of INT12 and GRAY8. Wall et al. (2004) reported similar coefficients of determination ($R^2 = 0.39 - 0.43$) for models which included averaged value of four independent ultrasound measurements at site between 12th and 13th rib. Aass et al. (2009) reported higher coefficients of determination ($R^2 = 0.80$) of models for intramuscular fat content prediction.

Acknowledgement

This article was written during realization of the project "CEGEZ No. 26220120042" supported by the Operational Programme Research and Development funded from the European Regional Development Fund.

References

Aass, L., Gresham, J. D., Klementsdal, G. 2006. Prediction of intramuscular fat by ultrasound in lean cattle. *Livestock Sci.*, 101, 228 – 241.

Aass, L., Fristedt, C.-G., Gresham, J. D. 2009. Ultrasound prediction of intramuscular fat in lean cattle. *Livestock Sci.*, 125, 177-186.

Albrecht, E. et al. 2006. Growth- and breed-related changes of marbling characteristics in cattle. J. Anim. Sci., 84, 1067-1075.

Brethour, J. R. 1994. Estimating marbling score in live cattle from ultrasound images using pattern recognition and neural network procedures. *J. Anim. Sci.*, 72, 1425 – 1432.

Faucitano, L. et al. 2004. Distribution of intramuscular fat content and marbling within the longissimus muscle of pigs. *Can. J. Anim. Sci.*, 84, 57 - 61.

Hassen, A. et al. 1999. Repetability of ultrasound-predicted percentage of intramuscular fat in feedlot cattle. *J. Anim. Sci.*, 77, 1335 – 1340.

Hassen et al. 2001. Predicting percentage of intramuscular fat using two types of real-time ultrasound equipment. J. Anim. Sci., 79, 11 – 18.

Herring, W. O. et al. 1998. Comparison of four real-time ultrasound systems that predict intramsucular fat in beef cattle. *J. Anim. Sci.*, 76, 364 – 370.

Indurain, G. et al. 2006. Composition and estimation of intramuscular and subcutaneous fatty acid composition in Spanish young bulls. *Meat Sci.*, 73, 326 – 334.

LUCIA – Users Guide, System for Image Processing and Analysis. Praha : Laboratory Imaging, CR, 2005.

Polák, P. et al. 2008. Prediction of intramuscular fat in live bulls using real-time ultrasound and image analysis. *J. Anim. And Feed Sci.*, 17, 32 – 42.

Reverter, A. et al. 2000. Genetic analyses of live-animal ultrasound and abattoir carcass traits in Australian Angus and Hereford cattle. *J. Anim. Sci.*, 78, 1786 – 1795.

SAS, 2005. SAS Users Guide. Version 9.1. SAS Institute Inc., Cary, N.C., USA.

Tomka, J. et al. 2007. Relationship between in vivo predicted and laboratory determined intramuscular fat content in bulls of differen breeds. *Slovak J. Anim. Sci.*, 40, 121 – 125.

Wall, P. B. et al. 2004. Use of ultrasound to predict body composition changes in steers at 100 and 65 days before slaughter. *J. Anim. Sci.*, 82, 1621 – 1629.