Effect of different ultrasound intensity levels on accuracy of intramuscular fat prediction in live pigs

J. Tomka, I. Bahelka, M. Oravcová, D. Peškovičová, E. Hanusová, R. Lahučký, P. Demo
Slovak Agricultural Research Centre – Research Institute for Animal Production, 949 92 Nitra, Slovak Republic

Summary

The objective of this study was to evaluate the possibility of prediction of intramuscular fat (IMF) in live pigs using ultrasound method. The accuracy of prediction at five different ultrasound intensity levels was investigated. Cross-sectional images of longissimus dorsi muscle (LD) at right last rib area from hybrid pigs were taken. Each pig was scanned at the same frequency (3.5 MHz) and at the five different ultrasound intensity levels. The video image analysis was used to predict IMF content (UIMF70 to UIMF90). A sample of LD at the last rib was taken for laboratory analysis of IMF content (LAIMF). Correlations between LAIMF and UIMF were significanly different from zero (r=0.40-0.52), except for correlation between LAIMF and UIMF90 (r=0.14). Statistical model with LAIMF the dependent variable, UIMF and live weight the covariates, and sex the fixed effect was developed. Coefficients of determination (R^2) were 0.33, 0.38, 0.34, 0.25 and 0.17 with UIMF at the intensity level 70, 75, 80, 85 and 90%. Root mean square errors (RMSE) ranged from 0.516 to 0.639%. Standard errors of individual prediction (SEP) ranged from 0.523 to 0.649%. Goodness-of-fit of the model was also justified by testing the residuals for normality. Although the results are not quite unequivocal in favour of the one intensity level, it seems that intensity levels 75 and 80% are the most suitable to predict IMF in live pigs. Further research is needed, mainly to increase accuracy of collecting, processing and evaluating the sonograms using video image analysis.

Key words: pig, intramuscular fat, prediction, ultrasound intensity level, correlations

Introduction

At present, breeding goals and strategies in pig husbandry shift from the quantitative parameters (percentage of lean meat, average daily gain, feed consumption, backfat thickness etc.) to traits of meat quality. Fresh pork quality has become important and has received more attention as producers and processors try to meet consumer demand for high quality pork. Drip loss, tenderness and intramuscular fat are regarded the most important parameters to take into account in order to assess the quality of pork (Monin, 1998; Eggert *et al.*, 2002; Mörlein *et al.*, 2005). It is generally accepted that an increased level of intramuscular fat has a positive influence on the sensory qualities of pig meat (Fernandez *et al.*, 1999). Greater amounts of intramuscular fat have been shown to positively impact sensory panel traits such as tenderness, juiciness, and flavour, along with mechanical measures of tenderness (Hodgson *et al.*, 1991; NPPC, 1995; Huff-Lonergan *et al.*, 2002). Heritability estimates for pork quality traits are low with exception of intramuscular fat which has moderate up to high values from 0.29 to 0.81 (Cameron, 1990; de Vries *et al.*, 1994; Knapp *et al.*, 1997; Liu *et al.*, 1998). Therefore, the effective selection for this trait is possible.

The aim of this study was to assess the possibility of prediction of intramuscular fat in live pigs using ultrasound method, and to compare the accuracy of prediction.

Material and methods

Data were collected from 144 hybrid pigs, which were progeny of White Meaty sows mated Hampshire x Pietrain boars. Pigs were weighed and measured one-three days prior to slaughter by ultrasound device ALOKA SSD-500 fitted with probe UST-5044-3.5 (3.5 MHz/172 mm). Cross-sectional images of longissimus dorsi muscle and subcutaneous fat overlying the loin muscle at right last rib area were taken. Each pig was scanned at the same ultrasound frequency (3.5 MHz) and at the five different ultrasound intensities: 70, 75, 80, 85, and 90% of total amplifying of sonograph (described as ultrasound intramuscular fat – UIMF70, UIMF75, UIMF80, UIMF85 and

UIMF90). The ultrasound measurements were performed by experienced technician. Vegetable oil was used as a conducting material between the probe and skin. At scanning, the echocoupler connected to probe was used to better adjusting to rounded contours of pig body and to capture whole muscle eye area. The ultrasonic images were digitized and stored in computer for evaluation.

The video image analysis was used to predict content of intramuscular fat (IMF). Software LUCIA (User´s guide, System for Image Processing and Analysis, Laboratory Imaging, Prague, Czech Republic, 2005) was applied. The method (peaks detection function) is based on enhancing small light objects. This function enables the specific segmentation of the small objects through their exclusion from the larger objects and can also help in the case of non-homogeneous background. Estimation of IMF content was calculated as proportion of IMF area to total marked loin muscle area in ultrasonic picture.

Pigs were slaughtered at 114.0±9.17kg live weight in experimental slaughterhouse of RIAP (Research Institute for Animal Production) in Nitra. The second day after slaughter, the dissection of right half carcass was done. A sample of longissimus dorsi muscle (approx. 100g) at the last rib (the same place as ultrasonic images were made) was taken for laboratory analysis of IMF content (LAIMF) using device INFRATEC 1265 Meat Analyzer (CM Instruments Laboratorgeräte GmbH, Bünde, Germany).

Statistical package SAS/STAT (2002-2003, v. 9.1.3) was employed in the analyses. Basic statistics was done using MEANS procedure. CORR procedure was used to calculate the Pearson´s correlation coefficients between UIMF70 to UIMF90 and LAIMF. The same statistical model (GLM procedure) was used to evaluate prediction ability of ultrasound IMF taken at various intensity levels:

$$y_{ij} = \mu + S_i + b_1 L_{ij} + b_2 W_{ij} + e_{ij}$$

where:

y_{ij} - individual observation of LAIMF (%)

μ - intercept

S_i – fixed effect of sex; $\sum_{i=1}^{2} S_i = 0$

L_{ij} – UIMF (%) taken at single intensity level (70, 75, 80, 85, 90%) i.e. UIMF70 to UIMF90

b_1 – linear regression coefficient of dependence of LAIMF on UIMF

W_{ij} – live weight (kg)

b_2 – linear regression coefficent of dependence of LAIMF on live weight

e_{ij} – random error $N\left(0, \sigma_e^2\right)$

To assess goodness-of-fit of the model, root mean square errors (RMSE) and standard errors of individual prediction (SEP) were calculated. The residuals were tested for normality (UNIVARIATE procedure).

Results and discussion

Basic statistics for carcass traits associated with animals in which the pairs of LAIMF and UIMF were available at different intensity levels (70, 75, 80, 85, 90%) is shown in Table 1. The values and standard deviations were almost the same.

Table 1 *Basic characteristics for carcass traits*

	Range		
Trait	Mean	S.D.	Min. – Max.
Carcass weight, kg	89.76 – 90.59	6.94 – 7.40	69.00 – 105.00
Backfat thickness, mm	27.36 – 27.93	5.08 – 5.35	16.67 – 42.00
Lean meat content, %	55.15 – 55.63	4.24 – 4.38	45.63 – 66.37

Basic statistics for UIMF taken at various intensity levels (UIMF70 to UIMF90) and LAIMF is shown in Table 2. The average UIMF increased with the intensity level applied and ranged from 1.22 to 3.41 %; the average LAIMF showed a stabile pattern with values from 2.10 to 2.29 %.

Table 2 *Basic statistics for ultrasound IMF taken at various intensity levels and laboratory analysed IMF (taken into account only measurements for which pairs of ultrasound and laboratory analysed values were available)*

Intensity level	UIMF (%)					LAIMF (%)			
	N	Mean	Median	S.D.	Range	Mean	Median	S.D.	Range
70 %	142	1.22	1.00	0.85	0.10 - 3.60	2.10	2.00	0.66	1.00 – 4.30
75 %	144	1.77	1.65	1.04	0.20 - 5.10	2.12	2.00	0.65	1.00 – 4.30
80 %	144	2.28	2.20	0.95	0.30 - 4.80	2.22	2.00	0.70	1.00 – 5.40
85 %	144	2.76	2.60	1.13	0.20 - 5.70	2.25	2.20	0.67	1.10 – 4.90
90 %	133	3.41	3.50	1.01	0.50 - 5.70	2.29	2.20	0.69	1.10 – 5.40

UIMF – ultrasound IMF at different intensity levels, LAIMF – laboratory analysed IMF

Pearson´s correlation coefficients between LAIMF and UIMF are presented in Table 3. Significant correlations were found between LAIMF and UIMF70 to UIMF85, except for UIMF90. The strongest relations were observed between LAIMF and UIMF75 and/or UIMF80.

Table 3 *Pearson´s correlation coefficients between ultrasound and laboratory analysed IMF*

Trait	UIMF75	UIMF80	UIMF85	UIMF90	LAIMF
UIMF70	0.62[a] (122)	0.46[a] (106)	0.30[b] (105)	0.22[c] (97)	**0.46**[a] (142)
UIMF75	-	0.55[a] (111)	0.51[a] (108)	0.40[a] (96)	**0.52**[a] (144)
UIMF80		-	0.47[a] (119)	0.32[b] (104)	**0.51**[a] (144)
UIMF85			-	0.46[a] (114)	**0.40**[a] (144)
UIMF90				-	**0.14** (133)

UIMF – ultrasound IMF at different intensity levels, LAIMF – laboratory analysed IMF

[a] *$P<0.001$*, [b] *$P<0.01$*, [c] *$P<0.05$;* Number of observations in parentheses

Goodness-of-fit of the model is shown in Table 4. The model explained 17 (UIMF90 covariate) to 34-38% (UIMF80 or UIMF75 covariate) of total variability of LAIMF. Root mean square errors (RMSE) ranged from 0.516 (UIMF75 covariate) to 0.639% (UIMF90 covariate). Standard errors of individual prediction (SEP) ranged from 0.519 to 0.536% (UIMF75 covariate) and from 0.644 to 0.667% (UIMF90 covariate).

Table 4 *Criteria to assess goodness-of-fit of the model*

Intensity level	R^2	RMSE for laboratory IMF (%)	SE of individual predictions for laboratory IMF (%)		Residuals range for laboratory IMF (%)
			Mean	Range	Observed - Predicted
70 %	0.33	0.546	0.553	0.549 – 0.568	〈-1.285; 1.719〉
75 %	0.38	0.516	0.523	0.519 – 0.536	〈-1.387; 1.360〉
80 %	0.34	0.576	0.584	0.580 – 0.599	〈-1.056; 2.425〉
85 %	0.25	0.586	0.594	0.590 – 0.608	〈-1.278; 1.957〉
90 %	0.17	0.639	0.649	0.644 – 0.667	〈-1.280; 2.803〉

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

Dion *et al.* (1996) predicted marbling score in pigs with real-time ultrasonic cross-sectional and longitudinal scans. The accuracy of prediction was essentially zero. In our study, correlations between LAIMF and UIMF ranged from 0.14 ($P>0.05$) to 0.52 ($P<0.001$). Similar correlations (r=0.46 to 0.60; r=0.50) between predicted IMF and that from carcass samples were reported by Newcom *et al.* (2002 and 2005). The higher correlations (r=0.70; r= 0.52 to 0.71) between analysed and predicted IMF were found by Ragland *et al.* (1997 and 1998). Bahelka *et al.* (2006 and 2007) reported correlations between LAIMF and UIMF at three different intensity levels ranging from 0.13 to 0.26 (frequency 5.0 MHz), and ranging from 0.13 to 0.31 and from 0.09 to 0.18 (frequency 3.5 and/or 5.0 MHz).

Coefficients of determination (R^2) ranged from 0.17 (UIMF90 covariate) to 0.38 (UIMF75 covariate). Except for intensity level 90%, the highest proportion of variability of LAIMF (about 2/3) separated through the model applied was due to differences in UIMF. The remaining proportion of variability was due to sex and live weight. When only UIMF was considered the independent variable, R^2 were lower and ranged from 0.15 to 0.28. The only exception was UIMF90 with $R^2<0.05$. R^2 between 0.33 and 0.38 were in agreement with findings of Ragland *et al.* (2002), who reported R^2: 0.33 to 0.38, of Newcom *et al.* (2002), who reported R^2 = 0.32 and of Leaflet *et al.*(2006), who reported R^2 = 0.36. Depending on hybrid combination evaluated and traits involved in the model equation (either linear or quadratic term), Eggert and Schinkel (1998) reported R^2 ranging from 0.50 to 0.83.

RMSE and SEP ranged from 0.516 to 0.639 and from 0.523 to 0.649 with the lowest values found when UIMF75 was considered the covariate. Ragland *et al.* (2002) found MSE in the range between 1.04 and 1.12 in dependence on prediction equations. The higher MSE (1.31 and 1.02 %) were reported by Leaflet et al. (2006) and Newcom *et al.* (2002). Newcom *et al.* (2002) and Ragland *et al.* (1997 and 2002) also reported the higher SEP: 0.80 to 0.93 %; 0.83 and 0.96 %.

The biggest problem to determine the accuracy of prediction is correct estimation of intramuscular fat content in ultrasonic images. In the muscles there are situated blood capillares which are considered by sonograph device as excessive marbling of muscle – so called „scattering effect“ (very bright spots in the screen). This effect is closely connected with intensity level and frequency of ultrasound. It is demonstrated at the higher frequency and intensity level. Using the lower frequency and intensity, „scattering effect“ is suppressed but it results in higher absorption of ultrasound and marbling is fading. This opinion is in agreement with Leaflet *et al.* (2006) who state that ultrasonic images are heavily influenced by system settings, technician experiences and various other conditions such as temperature, animal preparation, and electrical interference. Whittaker *et al.* (1992) also showed the scattering effect and absorption to be the main factors influencing interpretation of sonograms. The greater scattering effect in images of *longissimus dorsi* muscle is due to high frequency of probe used and very high ultrasound intensity applied. On the other hand, the lower ultrasound frequency and intensity applied, the higher absorption is.

The classification of pigs according to absolute values of residuals between observed and predicted IMF indicates the accuracy of prediction. 96 % of residuals (Int75 and Int80) were ≤ 1 %. Ragland (1998) reported predicted IMF within 1 % of carcass IMF in 72 to 86 % of the observations, depending on the model.

The results presented in this study document the ability to measure IMF in live pigs using ultrasound method. Although they are not quite unequivocal in favour of the one intensity level, it seems that intensity levels 75 and 80 % are the most suitable to predict IMF in live pigs. However, further research is needed, mainly to increase the accuracy of collecting, processing and evaluating the sonograms using video image analysis.

References

Bahelka, I. et al. 2006. Prediction of intramuscular fat proportion at pigs in vivo by ultrasound method. Conf. Biotechnology 2006, 15-16 February 2006, České Budějovice, CR, 141-143.

Bahelka, I. et al. 2007. The effects of probe type and intensity of ultrasound on accuracy of intramuscular fat prediction in longissimus dorsi muscle of pigs. *Biotechnology in Animal Husbandry*, 23, 87-95.

Cameron, N.D. 1990. Genetic and phenotypic parameters for carcass traits, meat and eating quality traits in pigs. *Livestock Production Science.*, 26, 119.
Dion, N.D. et al. 1996. Efficiency of various live measurements for the prediction of lean yield and marbling. Proc. Natl. Swine Improv. Fed. Mtg., Ontario, Canada.
Eggert, J.M. et al. 2002. Myosin heavy chain isomorphs account for variation in pork quality. *Meat Science*, 61, 117-126.
Eggert, J.M., Schinckel, A.P. 1998. Growth and characterization of individual backfat layer in swine: A new method for the prediction of intramuscular fat in the live animal or carcass. Record of Proceedings Nat. Swine Improvement Federation Conference and Animal Meeting. December 4-5, NSIF, vol. 23. East Lansing Marriot, Michigan. from www.nsif.com/Conferences /1998/Content/htm
Fernandez, X. et al. 1999. Influence of IMF content on the quality of pig meat – 1. Composition of the lipid fraction and sensory characteristics of m. longissimus lumborum. *Meat Science*, 53, 59-65.
Hodgson, R.R. et al. 1991. Relationship between pork loin palatability traits and physical characteristics of cooked chops. *Journal of Animal Science*, 69, 4858-4865.
Huff-Lonergan, E. et al. 2002. Correlations among selected pork quality traits. *Journal Animal Science*, 80, 617-627.
Knapp, P. et al. 1997. Genetic parameters for lean meat content and meat quality traits in different pig breeds. *Livestock Production Science*, 52, 69-73.
Laboratory Imaging, Prague,CR,2005. LUCIA – User´s guide, System for image processing and analysis.
Leaflet, A.S. et al. 2006. Development of a model to predict intramuscular fat in live pigs using real-time ultrasound. Iowa State University Animal Industry Report, from www.ans.iastate.edu/report/air/2006pdf/R2050.pdf
Liu, E.C. et al. 1998. Simultaneous estimation of genetic parameters for 38 traits of growth, carcass, meat quality and eating quality. *Joint Meet. Abstr*., 78.
Monin, G. 1998. Recent methods for predicting quality in whole meat. *Meat Science*, 49, p231-243.
Mörlein, D. et al. 2005. Non-destructive estimation of the intramuscular fat content of the longissimus muscle of pigs by means of spectral analysis of ultrasound echo signals. *Meat Science*, 69, 187-199.
Newcom, D.W. et al. 2002. Prediction of intramuscular fat percentage in live swine using real-time ultrasound. *Journal of Animal Science*, 80, 3046-3052.
Newcom, D.W. et al. 2005. Comparison of three models to estimate breeding values for percentage of loin intramuscular fat in Duroc swine. *Journal Animal Science*, 83, 750-756.
NPPC 1995. Genetic evaluation/Terminal line program results. R. Goodwin and S. Burroughs, ed. Natl. Pork Prod. Council, Des Moines, Iowa.
Ragland, K. et al. 1997. Prediction of intramuscular fat in live swine using real-time ultrasound. Rec. Proc. Natl. Swine Improv. Fed. Conf. Ann. Mtg., 5-6 December, Des Moines, Iowa, USA vol. 22.
Ragland, K.D. 1998. Assessment of intramuscular fat, lean growth, and lean composition using real-time ultrasound. Ph.D. Dissertation. Iowa State University, Ames.
Ragland, K.D. et al. 2002. Efficacy of using real-time ultrasound to predict intramuscular fat in the *longissimus* muscle of live swine. from www.ext.vt.edu/news/periodicals/livestock/apr02_12/aps-171.html
SAS/STAT 2002-2003. SAS Institute Inc., ver. 9.1.3, Cary, N.C., USA.
De Vries, A.G. et al. 1994. Genetic parameters of pork quality and production traits in Yorkshire populations. *Livestock Production Science*, 40, 277-289.
Whittaker, A.D. et al. 1992. Principles of ultrasound and measurement of intramuscular fat. *Journal of Animal Science*, 70, 3, 942-952.