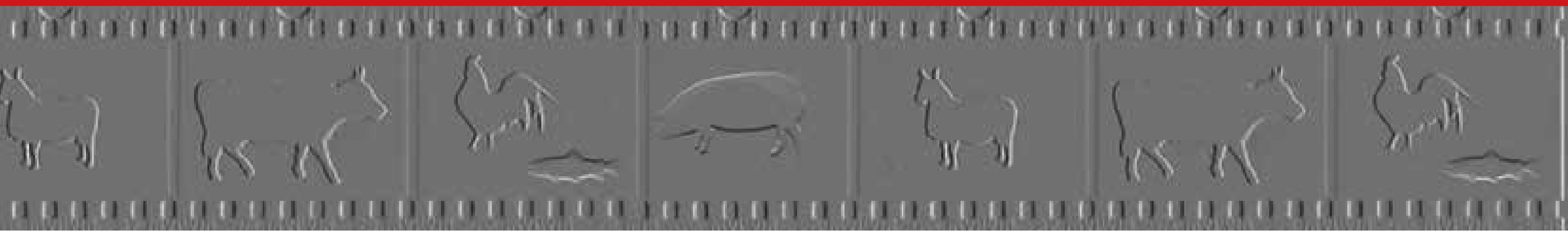
FAIM

Farm Animal IMaging

FARM ANIMAL IMAGING

KAPOSVÁR 2013



C. Maltin, C. Craigie and L. Bünger

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Introduction from the Editors

This book is mainly a report of the second annual conference of the COST action FA1102: FAIM which was held at Kaposvár in Hungary in October 2013. The major elements in the book are papers, which were presented at the FAIM II meeting in Hungary. Also included are the winners of the poster competition sponsored by EplusV.

The first prize poster is included as a paper, with the runners up having their posters included. The book also contains reports on the meeting itself, and some of the other activities carried out during the year by FAIM participants, including the EAAP conference in Nantes, a training school held in Rennes, and a letter of comments on the experience of undertaking a short term scientific mission (STSM).

The book also contains the outcome of a review to define standardized reference traits for the measurement of MQ criteria. This is a key task for workgroup 2 which was completed this year. We hope that you enjoy this book, and feel inspired to come and join us in FAIM.

To join please contact the action Chairman, Dr Lutz Bünger using the contact details below or via the website **www.cost-faim.eu**.

Dr Lutz Bünger
SRUC Easter Bush Estate,
Edinburgh Scotland
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Finally, on behalf of all FAIM participants, we wish to thank COST for funding the COST action FA1102 (FAIM) and related activities undertaken in 2012-2013.

Charlotte Maltin,
Cameron Craigie
and Lutz Bünger – editors.

A Report on FAIM at EAAP in August 2013 in Nantes, France

L. Bünger

The 64th Annual Meeting of the European Federation of Animal Science took place at the end of August in Nantes, France under the theme:

“New challenges facing animal production for diversified territories, market demands and social expectations”.

FAIM was given a special opportunity at this year’s EAAP conference recognising the reputation FAIM has built in its first year. As chair of the COST Action FAIM (*Optimising and standardising non-destructive imaging and spectroscopic methods to improve the determination of body composition and meat quality in farm animals*) Dr Lutz Bünger was invited by the French Organising Committee to chair one session at the EAAP with the theme: Carcass and meat quality: from measurement to payment. This exactly met the remit of FAIM and so this offer for one session evolved to a full one day symposium because we received well over 20 applications for talks and a similar number for poster presentations, and the FAIM presence was immense! This indicates the large interest in this subject which hopefully will be reflected in the upcoming EU Framework Programme for Research and Innovation: Horizon 2020.
23 talks were finally presented in the FAIM led symposium and 20 posters among them well known FAIM Members as invited speakers like Prof Armin Scholz who presented a paper entitled Non-invasive measurement of body and carcass composition in livestock by CT, DXA, MRI, and US.; Dr Gerard Daumas speaking on CT & Automatic Imaging Systems for a Value-Based Marketing System in Pigs, Prof Rainer Roehe presenting online techniques to measure meat quality and Dr Cameron Craigie from QMS reviewing the importance of video-image analysis for a value-based marketing system for beef and lamb.

Around 300 of the 1300 EAAP delegates attended talks in the FAIM led one-day-symposium. It was a long and very exciting day for all participants of which most were already members of the COST Action FAIM. On the day, more scientists joined the FAIM Action bringing the total FAIM membership to well over the 250 mark.

This exciting day finished with a FAIM meeting of WG1 and WG2 during which we summarised the symposium and discussed further preparations for our annual conference FAIM II (29/30 October 2013 in Kaposvár/Hungary) featuring 20 talks and 25 posters including a wine reception sponsored by EplusV.

Photo: Armin Scholz

Further discussion followed on the Training School in Rennes: **Pig carcass composition measurement by CT and MRI - Live Pig measurement by CT- From acquisition to data analysis**

FAIM brings together now over 275 experts from 22 (25) EU countries (and beyond). It aims to optimise non-destructive *in vivo* (iv) and post mortem (pm) imaging and spectroscopic methods for the measurement of body composition and meat quality (MQ) in major farm animal species and to devise standardised principles of carcass classification and grading across countries.

These actions are necessary for the development of value-based payment and marketing systems and to meet the urgent need for market orientated breeding programmes. FAIM encompasses collaboration of hard- and software manufacturers with livestock and imaging academic experts to develop required products for implementing the scientific work. FAIM will coordinate and strengthen EU scientific and technical research through improved cooperation and interactions.

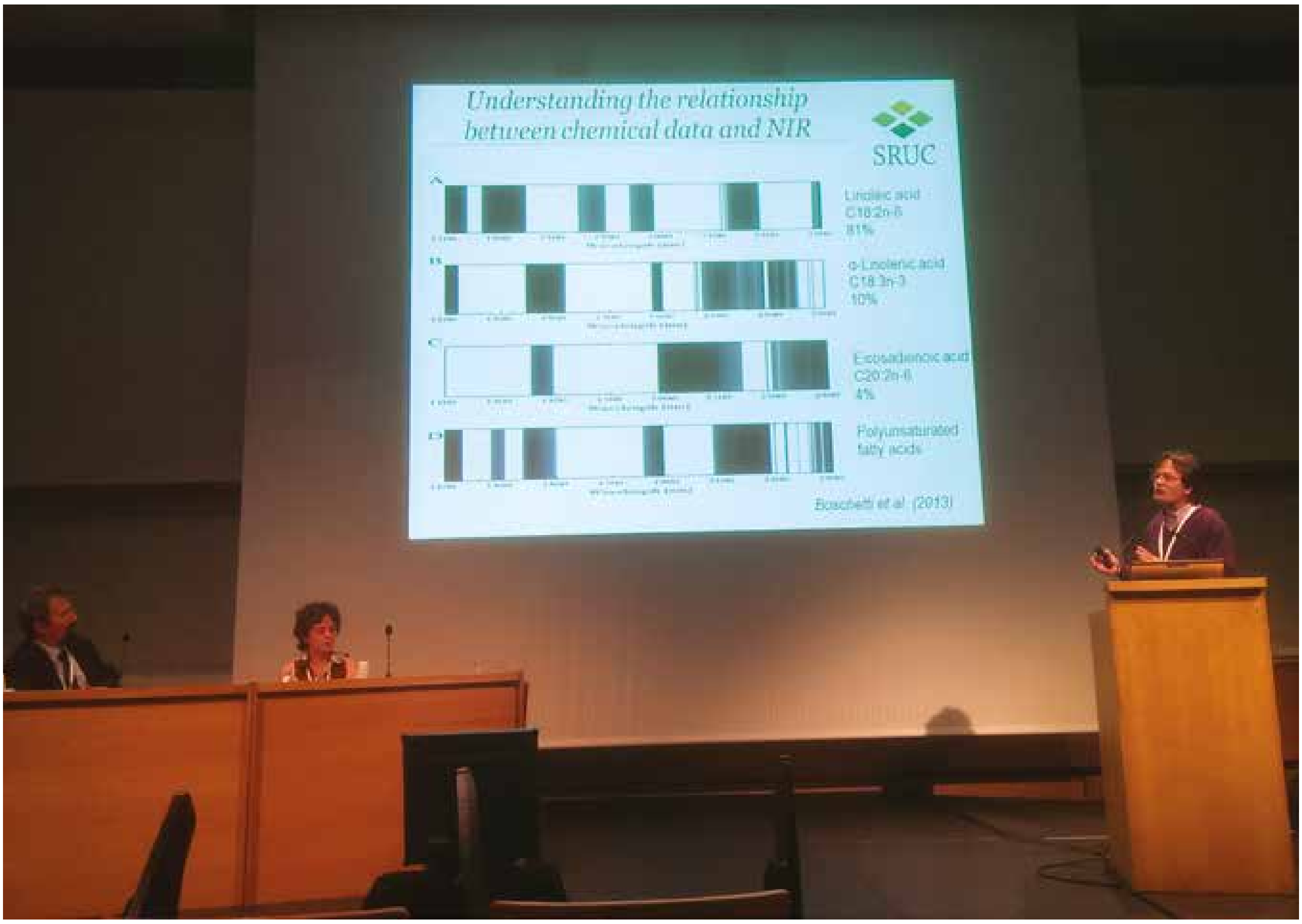


Figure 1. Rainer Roehe (SRUC) presenting his keynote talk at EAAP in Nantes during the one-day FAIM session ‘Carcass and meat quality: from measurement to payment’.

Training School at Rennes - Report

Pig carcass composition measurement by CT and MRI, Living pig measurement by CT - from acquisition to data analysis.

This training school was organized by Mathieu Monziols and Gerard Daumas from IFIP, Rennes and was held between the 8th and 10th of October 2013. The objective was to share best practice techniques for image acquisition and analysis from live pigs, carcasses and joints with the participants. Twelve participants, four trainers and two technicians from 11 different countries attended the training school (a list of attendees is appended).

All the participants and trainers agreed that the training school was very useful and successful. The participants asked many questions indicating large interest they had in the training school. Participants who came from different countries with different experiences shared and exchanged their knowledge and technical problems with the trainers.

So, the training school opened new opportunities for technology transfer and data exchange. Finally, the participants strongly supported the importance of conducting and continuing such training schools in future perhaps on an annual basis, while the trainers expressed their continuous support to the participants in their current and future research.

The organizers would like to thank COST FAIM for funding this training school, the travel and subsistence for participants and all the people that took part in organizing these very fruitful days.

The programme for the training school is outlined below:

Tuesday 8th October

Morning (IFIP Le Rheu)

- Trainees welcome (Mathieu Monziols and Gerard Daumas)
- CT and MRI imaging techniques presentation (Mathieu Monziols)
- CT at IFIP presentation (Mathieu Monziols)
- MRI at IRSTEA presentation (Mathieu Monziols)
- CT at SRUC presentation (Lutz Bünger)

Afternoon (IFIP Experimental station, Romillé)

- EU carcass preparation demo (Mathieu Monziols and Eric Gault)
- Carcass CT scanning practice (Mathieu Monziols and Eric Gault)

Figure 1. CT scanning of half pig carcass.

Wednesday 9th October

Morning: IRSTEA Rennes

- MRI carcass joints scanning (Mathieu Monziols and Stephane Quellec)

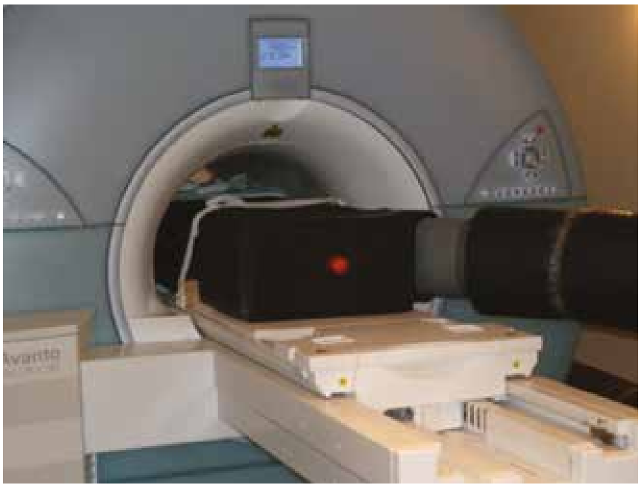


Figure 2. MRI scanning a loin.

Afternoon: IFIP experimental station, Romillé

- Anesthesia presentation and demo (Anne Hemonic and Eric Gault)
- Live animal CT scanning demo (Mathieu Monziols and Eric Gault)

Thursday 10th October

Morning: IFIP Le RHEU / Romillé

- Image analysis generalities (Mathieu Monziols)
- Image analysis thresholding practice with Image J (Mathieu Monziols)
- Semi-automatic Image analysis demo on live animal for viscera separation (Mathieu Monziols)
- Automatic image analysis demo for body composition (Mathieu Monziols)

Afternoon: IFIP Le RHEU

- Principles of statistical analysis (Gerard Daumas)

Figure 3. CT scanning a live pig

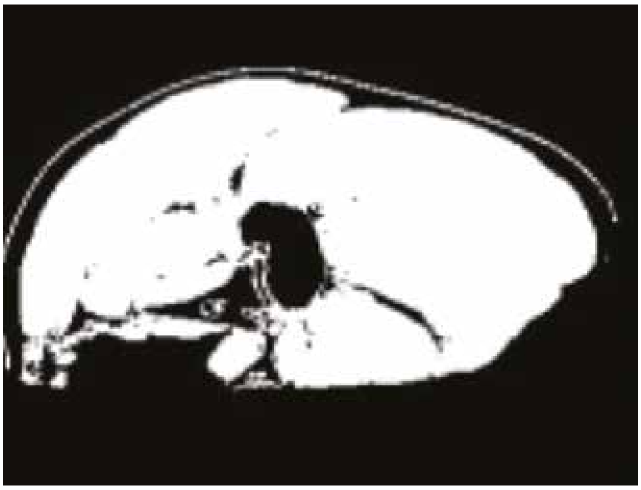


Figure 4. Image thresholding is used to identify muscle, bone and fat

Participants:

Georgios Arsenos: University of Thessaloniki, Greece
Khalfan Mohamed Al-Rashdi: University of Stirling, UK
Albert Brun: IRTA, Spain
Tamas Donko: Kaposvár University, Hungary
Lars Erik Gangsei: Animalia, Norway
Beata Grzegrzolka: Warsaw University, Poland
Stijn Hellebuyck: Ghent University, Belgium
Michael Judas: MRI, Germany
Jørgen Kongsro: Norsvin, Norway
Agnieszka Ludwiczak: Poznan University, Poland
Daiva Ribikauskiene: Lithuanian University of Health Sciences, Lithuania
Vanessa Salvi: Teagasc, Ireland

Trainers:

Lutz Bünger: SRUC, Scotland
Gerard Daumas: IFIP, France
Anne Hemonic: IFIP, France
Mathieu Monziols: IFIP, France

Technical support:

Eric Gault: IFIP, France
Stephane Quellec: IRSTEA, France

Figure 5. Pig holding pen

Figure 6. Attendees at the Rennes Training School

Short term Scientific mission

C.R.P.A.
Centro Ricerche Produzioni Animali - C.R.P.A. S.p.A.
Sede legale: Corso Garibaldi, 42 - 42121 Reggio Emilia – I
Sede operativa: Viale Timavo,43/2 - 42121 Reggio Emilia – I
Tel. +39.0522.436999 - Fax +39.0522.435142
www.crpa.it - info@crpa.it – crpa@postacert.vodafone .it
Part. IVA 01253030355 - R.E.A. 199780
C.F. 80010710350 - Cap.Soc. 1.851.350,00 €

Reggio Emilia, September 25th, 2013

***Subject**: STSM*

Dear Prisca,

My experience was at IFIP institute du porc in Rennes in France.

It is the most important institute responsible of the French pig population grading, in particular regarding the use of CT scanning technology for the determination of pork quality.

In this period I have learnt more about CT technology use and its application on pig carcass.

Furthermore, I have taken contact with people involved in pig grading work by Image Meater equipment and with the main classification organization (Uniporc Ouest).

This was very interesting for my job in Italy, because the possibility of applying of this work philosophy in my Country.

I had the possibility to visit the annual Fair on Animal Production (SPACE) too and to meet and to discuss with a lot of pig organizations.

In conclusion my experience was very positive because I have learnt important things for my pig career.

Andrea Rossi

Figure 1. CT Scanning pork joints

About short term scientific missions

The aim of a Short-Term Scientific Mission (STSM) is to contribute to the scientific objectives of the COST Action. These Missions (Exchange Visits) are aimed at strengthening the existing networks by allowing scientists to go to an institution or laboratory in another COST member state to foster collaboration, to learn a new technique or to take measurements using instruments and/or methods not available in their own institution/laboratory. They are particularly intended for young scientists. Further information regarding the application process for an STSM can be obtained from the action chairman Dr. Lutz Bünger by emailing **lutz.bunger@sruc.ac.uk.**

FAIM II – KAPOSVÁR Meeting report

T.Donkó and G.Milisits

Kaposvár University, Hungary

The COST FAIM II conference for 2013 was held in Hungary and was kindly hosted by Tamás Donkó and Gábor Milisits from the Kaposvár University.

Figure 1. Conference delegates during the conference.

Kaposvár University was established in its current form in 2000, and has a modern well equipped campus suitable for organizing such a conference.

Prior to the start of the conference a number of the conference participants were treated to a visit to the University's study farm and a trip around the excellent facilities of the Faculty of Agricultural and Environmental Sciences. During the visit the group was treated to a tractor trip round the deer farm and an excellent lunch.

The study farm covers some 1360 hectares supporting the raising of all major farm animal species, a game farming centre (deer and wild boar) and serves both as a farm and a visitor centre. The visitor centre demonstrates a range of livestock, a game farming museum and caters for the visitors with an onsite restaurant.

Figure 2. A visit to the Kaposvár University farm.

Figure 3. The Hungarians looked after their guests well.

The participants also visited the Faculty's well equipped laboratories and an excellent imaging centre located at the main campus of Kaposvar University. These included the Department of Agricultural Product Processing and Department of Food Development and Bioanalytics laboratories are equipped with NIR spectrometer, electronic nose and tongue, GC-MS, LC-MS, GC-FID, AAS and conventional meat quality technologies. The Institute of Diagnostic Imaging and Radiation Oncology is equipped with two CT scanners for human clinical imaging (Siemens Definition Flash and Siemens Somatom Emotion 6) and one dedicated CT scanner for non-human research activities (Somatom Sensation 16 Cardiac). Furthermore it has two MRI scanners (Siemens Magnetom Avanto 1.5T, GE Ovation Signa 0.35T) and a DSA laboratory.

The scientific aspects of the conference were held over two days, with day one being occupied by scientific presentations from the four working groups and day two being taken up with constructive debate and discussion on the key objectives of the COST action. The 100 participants came from 23 different countries (see the table opposite).

Figure 4. A tractor tour of Kaposvár University farm.

The key scientific areas covered in the conference were:

- The use of CT as a reference method for determination of body composition in pigs.
- The use of phantoms for standardisation of CT scanners.
- The urgent need for means for large animal imaging.
- A review of the spectroscopic and imaging technologies for determining meat quality.
- The role of CT in assessing intramuscular fat.
- The use of CT to drive robotic carcass cutting.
- Methods and algorithms for analysing images.
- The challenges and opportunities for electronic tagging.

Country	Persons
Australia	1
Belgium	3
Canada	1
Denmark	7
Spain	10
France	2
Germany	6
Greece	1
Hungary	22
Ireland	2
Island	2
Italy	5
Lithuania	1
Norway	4
New Zealand	2
Poland	1
Portugal	1
Slovenia	2
Serbia	2
Switzerland	3
Slovakia	3
Sweden	2
United Kingdom	17
23	**100**

Figure 5. Edwina Toohey from Australia during discussion.

Figure 6. Lutz discusses with Odd Vangen and Peter Horn.

Figure 7. Introduction to the farm visit.

Figure 8. Harvey Ho (ABI, New Zealand).

Figure 9. A demonstration of imaging at Kaposvár University.

Figure 10. Snapshot during the discussion.

Figure 11. Lutz and Gerard in discussion.

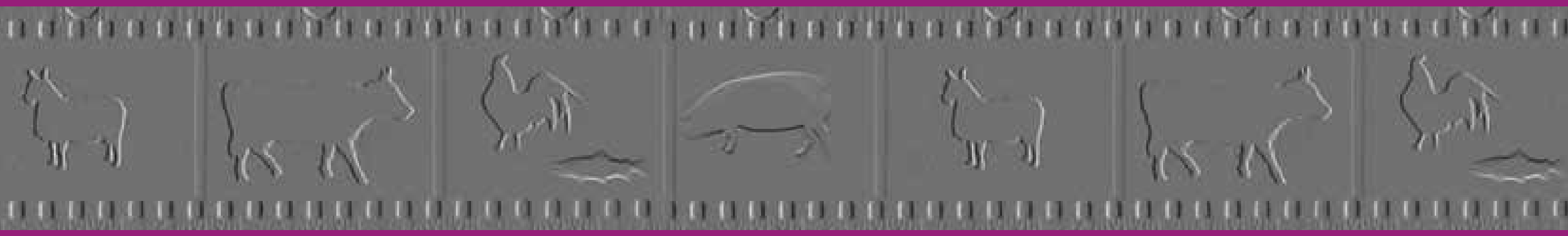
Figure 12. The reception desk was always busy.

Figure 13. Kaposvár University farm has impressive Red Deer and interesting cattle.

Figure 14. Lutz photographed by Beata during the farm visit.

Workgroup 1

Gerard Daumas
Eli Olsen
Mathieu Monziols
Lars Christensen
Torunn Aasmunstad
Anna Carabús



Identification of possible and relevant post mortem reference methods for carcass composition

G. Daumas[1], T. Donko[2] and M. Monziols[1]

1. IFIP-Institut du Porc, Le Rheu, France.
2. Kaposvár University, Kaposvár, Hungary.

Value for Industry

- CT can provide accurate measurements of carcass and body composition of the farm animals.
- CT has a recognized potential to be a primary reference, in particular for breeding purposes.
- Building an international CT based reference would improve the comparisons, the market and the efficiency of the whole chains.

Background

CT has been used as a working standard to measure body composition for many years by a lot of researchers. In particular, this has been very efficient in sheep breeding. Ten years ago, the EUPIGCLASS project recommended the introduction of CT as a potential reference in the EU regulation for pig grading. This was a starting point of new research in this area. Several countries developed their own working standard. However, without harmonization, CT cannot be used as a stand-alone (primary) reference; costly dissections still have to be done.

To build a primary CT reference for measuring body/carcass composition is one of the main aims of COST Action FAIM. To identify possible and relevant post mortem reference methods for carcass composition is one of the milestones for FAIM in 2013.

Few methods have been proposed where CT is considered as a primary reference, i.e. without any calibration against dissection. A procedure must contain acquisition parameters and image analysis, this latter seems to have the greatest impact.

Why work is needed

In order to identify possible reference post mortem (pm) it is important to review the CT methods used for carcass composition. This is necessary to clarify the content of the CT classes and the methods for the conversion of volumes into weights.
Moreover, in order to agree on a reference, the stakeholders need to know the impact of each parameter and in particular of those having the greatest impact. It seemed thus very important to assess the effect of tissues segmentation and the effect of conversion of volumes into weights.
Finally, focusing on pig carcass composition was quite obvious, as an urgent matter under discussion at a EU level and as the segmentation is more complex due to the presence of rind. This work is an important step in order to be able to propose future coordinated research works.

The methods used

First of all, an extended literature review on CT used for measuring compositional traits was performed. Articles using CT procedures stemming from calibrations (always against dissection) were excluded. Only the articles considering CT as a primary (stand-alone) reference were taken under consideration.

Our investigation was focused on the image analysis which is considered as the main source of differences. The scope was limited to muscle segmentation by thresholding, one of the most common method, one of the simplest ones, and a good candidate for a reference. Muscle Hounsfield range and muscle density were collected for each selected article.

An arbitrary reference was chosen among these ranges and densities to facilitate comparisons. The reference was decided by the authors after taking into account the more convincing arguments.

In order to assess the effect size of HU ranges and densities, we had to use some available datasets. We chose 3 recent trials involving extreme pig carcasses or pig joints from French and Italian pigs:

• Trial 1:
Calibration of the French pig classification methods, carried out in 2012. It involved 250 pigs representative of the French population, including females and castrated males. The 4 main EU joints were CT scanned.
• Trial 2:
Experiment in course on pig genomics, including in particular Pietrain entire males. The present dataset included about 1500 pigs. Carcass side was CT scanned.
• Trial 3:
Calibration of the Italian pig classification methods, carried out in 2012. It involved:
- 150 heavy pigs
(carcass weight in the range 115-150 kg),
- 150 light pigs
(carcass weight in the range 95-110 kg).
Following Italian jointing, the 5 main joints were CT scanned.

In the 3 trials the scans were performed with the same CT scanner, the mobile IFIP CT scanner (Siemens Emotion Duo), using the same acquisition parameters: 130 kV, 40 mAs, 3 mm slice thickness, spiral scanning, FoV 500x500 mm, acquisition matrix 512x512, reconstruction filter B30S (soft tissues). Trial 3 was considered as two datasets, one for each subpopulation.

Three carcasses were selected in each of the 4 datasets on the LMP basis: the lowest LMP, the highest LMP and the LMP closest to average. For this study the sample gathered 12 pigs with a huge variability.

The number of voxels was calculated within the muscle HU range of each selected publication in each of the 12 pigs. Then, the relative difference of muscle volume was calculated with the HU range [0-120], chosen as arbitrary reference (Daumas & al., 2011). For the publications having mentioned a muscle density, this density was applied to the muscle volume to calculate the relative difference of both muscle weight and LMP with the arbitrary reference (density = 1.04; ICRU (1989). Finally, for each article was calculated the extreme relative differences as well as the median difference, both for volume and LMP.

The results obtained

From the studied literature 15 references matched the study constraints. They concerned beef, pig, lamb and sheep, and *in vivo,* carcass or cuts. These 15 references used 11 muscle HU range. Several authors used the same muscle HU ranges. Table 1 summarizes the median relative differences with the study reference of CT muscle volume, sorted by ascending order, for the 11 muscle HU ranges (codified from 1 to 11). The study reference has the code 8. The range [0-120] was proposed as a reference by Daumas and Monziols in 2011 for pig carcasses. It was used too by Brun *et al.* (2012) on beef cuts.

Most of the median relative differences ranged between -9.4 % and +6.6 %. More extreme differences were obtained for Picouet *et al.* (2010) with -13% on hams and -21% on pig carcasses.

The inferior HU muscle limit comprised values between -22 and +30, while the superior limit comprised values between +76 and +200. The extreme values were thus -22 and +200, but the maximum range width was 201 HU, corresponding to the [0-200] range. The minimum range width was 54 HU, corresponding to the [23-76] range.

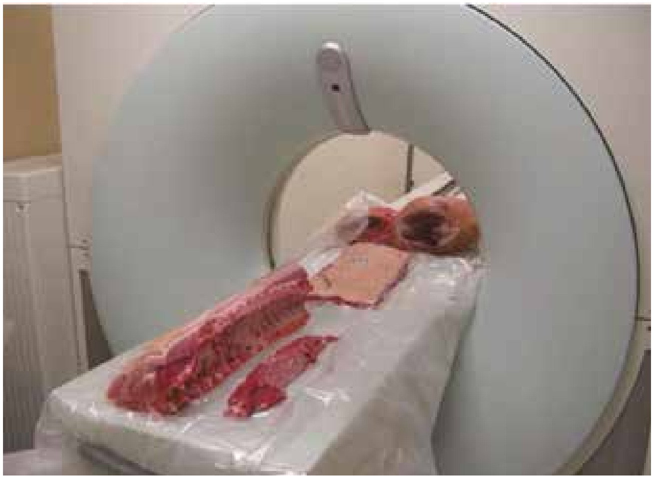


Figure 1. CT scanning the 4 main EU joints.

Table 1. Median relative differences with the study reference of CT muscle volume, sorted by ascending order, for the 11 muscle HU ranges (codified from 1 to 11), corresponding to the first author having used such a range.

				MUSCLE		Median difference
Code	**Authors**	**Species**	**Entity**	**Inf**	**Sup**	**Volumes**
1	Picouet *et al.* (2010)	Pig	Carcass	23	76	-21,1%
2	Picouet *et al.* (2010)	Pig	Ham	23	85	-12,7%
3	Navajas *et al.* (2010)	Beef	Cuts	30	133	-9,4%
4	Picouet *et al.* (2010)	Pig	Loin	7	85	-7,7%
5	Romvari *et al.* (2005)	Pig		20	200	-4,0%
6	Horn (1995)			10	150	-1,5%
7	Navajas *et al.* (2006)	Sheep	Live	-10	93	-0,5%
8	Daumas and Monziols (2011)	Pig	Carcass	0	120	0,0%
9	Monziols and Daumas (2010)	Pig	Carcass	0	200	1,8%
10	Campbell *et al.* (2003)	Sheep	Live	-17	120	4,3%
11	Kvame *et al.* (2004)	Lamb	Cuts	-22	146	6,6%

None of the authors converted the CT volumes into weights. Among the 15 articles, only 4 applied a muscle density (Md), using the 3 following references:

- Md = 1.04, citing ICRU (1989)
- Md = (HU x 0.00106) + 1.0062, citing Campbell *et al.* (2003), who cited Fullerton (1980).
- Md = (HU x 0.001413) + 0.997649, developed by Picouet *et al.* (2010).

The two last densities were a linear function of the HU values and were applied either to the average HU in the muscle range or by HU value. The latter method was applied in this comparison study. For 10 HU the variation of density was about 0.01 for both formulas. At 60 HU the "Campbell formula" gave a density of about 1.07 while the "Picouet formula" gave a density of about 1.09. Compared with the reference density of 1.04, differences are respectively of 0.03 and 0.05. For the same muscle volume, relative differences are thus about 3% and 5% respectively. Differences are lower at 50 HU.

The median relative difference of muscle weight and LMP ranged from -18 % to 9 %. The effect of using the "Campbell density" increased the relative difference of muscle volume of about 2.5 %. The effect of the "Picouet density" decreased the relative difference of muscle weight and LMP of about 3-3.5 %, compensating partially the lowest volume.

Table 2. Median relative differences with the study reference of muscle weight (and LMP), sorted by ascending order, with the codes corresponding to table 1.

					MUSCLE		Median difference	
Code	**Authors**	**Density used**	**Species**	**Entity**	**Inf**	**Sup**	**Volumes**	**Weights**
1	Picouet *et al.*(2010)	Picouet *et al.* (2010)	Pig	Carcass	23	76	-21,1%	-18,2%
2	Picouet *et al.* (2010)	Picouet *et al.*(2010)	Pig	Ham	23	85	-12,7%	-9,2%
4	Picouet *et al.* (2010)	Picouet *et al.* (2010)	Pig	Loin	7	85	-7,7%	-4,4%
8	Daumas and Monziols (2011)	ICRU (1989)	Pig	Carcass	0	120	0,0%	0,0%
9	Monziols and Daumas (2010)	ICRU (1989)	Pig	Carcass	0	200	1,8%	1,8%
11	Kvame *et al.* (2004)	Campbell *et al.* (2003)	Lamb	Cuts	-22	146	6,6%	9,0%

The scientific conclusions

Very few possible post mortem (primary) reference methods for carcass composition were identified in our literature review. We restricted this review to CT scanner and muscle volume measured by thresholding. All the authors thresholded CT scans into only 3 components: muscle, fat and bone. In pig carcasses, another tissue is present: the rind, which represents approximately 5% of the weight. Rind density is close to muscle density. Some specific approach to remove rind should be considered. Some authors applied mathematical morphology.

The inferior limit of muscle volume has the greatest impact because of a noticeable proportion of mixed voxels around 0: most of these mixed voxels correspond to a mix between muscle and fat tissues. Positive limits ranged between 0 and 30. Negative limits ranged between 0 and -22. An inferior limit comprised between -20 and +20 seems a good starting point for a relevant reference.

The superior limit of muscle volume has a lowest impact, unless this limit is very low. A limit less than 100 HU should underestimate muscle volume. A high upper limit should only slightly overestimate muscle volume, because of a low proportion of bones and mixed voxels between muscle and bones in this area. For instance, a 200 HU limit only increased the muscle volume of about 2% compared to a 120 HU limit when applied on this study sample.

We only found 3 muscle density values, established by ICRU (1989), cited by Campbell *et al.* (2003) and developed by Picouet *et al.* (2010). The first one was constant (1.04) while the two others were a linear function of HU. The differences between the three densities decreased as the HU decreased. At 60 HU, a value close to muscle peak on pig carcasses, the differences with 1.04 was 3% for Campbell *et al.* (2003) and 5% for Picouet *et al.* (2010). Such high differences should motivate future investigations on the variability of muscle density. Applying a density function of HU to a HU frequency histogram seems appealing. Nevertheless, this should be decided in accordance with the muscle thresholds and the method to manage the partial voxels.

The next steps

Further studies on the analysis of CT images should investigate:

- **the variability of tissues density,**
- **how to segment the rind when present,**
- **how to manage the partial volumes, especially between muscle and fat.**

Further studies should assess the impact of CT scanners parameters. A harmonized procedure of CT scanning and of analyzing CT images should be proposed for measuring carcass composition. Then, the accuracy of this proposed CT reference should be documented.

Acknowledgements

This work has been done thanks to a financial support from Inaporc and FranceAgriMer.

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Comparison of accuracy of reference methods based on CT and manual dissection

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Value for Industry

- Harmonization of instrumental online classification methods within the EU improves transparency of trade between member states. A precondition is a common reference method, which is considered to be the lean meat content expressed as a percent of the total weight, LMP.
- The classification results reflect the value of European pig populations. Homogeneous and, in most cases, lean pigs are generally preferred, and thus widely developed over the last decades. Further improvement requires better accuracy of both reference and online methods for measuring LMP. Online classification instruments are provided by commercial companies whereas the reference method is defined and approved by EU. The calibration of the commercial instruments is often carried out by public institutions according to the formalities required by the EU. Attention on cost and accuracy has resulted in the use of computed tomography, CT, as new reference method.
- Reference methods based on CT will potentially reduce costs and improve the accuracy of the reference method. Furthermore, slaughterhouses can define their own quality traits aimed at the production planning like the amount of ham or the quality of belly. Most of these traits can easily be obtained from the CT measurements used for the official reference method.

Background

The harmonized market and specific the need for transparency lead to regulations for objective classification in 1984/1985 laid down by the EU Council and Commission (latest revision COMMISSION REGULATION (EC) No 1249/2008 of 10 December 2008). The regulations do not limit the use of technology but define requirements on the predictive capability of the methods, evaluated by the quality of calibration. Statistical guidelines (Causeur *et al*. 2003) have been made, which propose cost effective solutions to practical challenges related to calibration, including sampling of the population.

Objective online classification of pig carcasses is obtained by measuring relevant characteristics highly correlated to the total lean meat content, LMP (Olsen *et al*. 2007). In most cases a linear transformation converts the measurements to a predicted value of LMP. The parameters of transformation are obtained using coherent data from both online measurements and a reference method.

The formal definition of the reference method for classification of pigs is total dissection of the prepared, half carcass. It is defined by the ratio between the total weight of lean meat in the carcass separated with a knife and the total weight of the carcass. However, a simplified method is possible defined by the ratio of the weight of lean meat in four main cuts of the carcass together with the tenderloin and the total weight of the same four cuts and the tenderloin. The ratio is multiplied with a factor (of 0.89) to obtain accordance with total dissection. Recently it has been approved to use computed tomography, CT, provided that an acceptable correlation with knife dissection methods can be demonstrated (Dobrowolski *et al* (2004), Romvari *et al* (2006), Vester-Christensen *et al* (2009), Font-i-Furnols (2009), Daumas *et al* (2011). No formal requirements are issued explaining the meaning of “acceptable correlation”.

Why work is needed

The idea of using objective classification to support transparency and a harmonized market is basically the right thing to do. However, the "objective classification" is not completely objective. It is dependent on the calibration method, the sample of data – and the people carrying out the calibration experiment. In our experience, the people involved in generating the data contribute significantly to the uncertainty, not deliberately, but because of the variation between individuals. Consequently, a reference method based on CT with minimal human impact on the measurement would be preferable. If precautions are not taken this is, however, only true with respect to repeatability. If the measuring protocol is not standardized to a certain extent, the variability may still be considerable. In this paper we draw attention to the metrological problems especially the identification of the critical factors that should be standardized to improve the robustness and reliability of the reference system for online classification of pig carcasses.

The method used

A budget of uncertainty of the reference methods for online classification is carried out using available uncertainty estimates from both published and un-published experiments. Uncertainty is estimated using two types of variance estimates, see the Guide to the expression of uncertainty in measurement, (GUM (2008). The type A uncertainty estimate is obtained from the experimental variance of observations, which typically are considered as outcomes from a Gauss distribution. The type B uncertainty estimate is evaluated by scientific judgment based on available information. If the range of outcomes can be determined without any knowledge of the distribution, the uniform distribution is assumed together with the variance estimate $a^2/3$, where "a" denotes the half range.

The results obtained

The primary standard for the content of lean meat in a pig carcass is defined as the ratio, LMP_0, between the weight of "red striated muscles from all parts (except the head) of the carcass as far as separable by knife" and the weight of carcass.

$$LMP_0 = \frac{\textit{Lean weight}}{\textit{Carcase weight}} \text{ x } 100\%$$

Transforming the definition into praxis rises some questions like: How should a carcass be defined? Can we assume a symmetric carcass and only dissect one half carcass? Is "as far as separable by knife" an unambiguous instruction? And do tendons, glands and blood vessels belong to lean meat?

The regulation includes some of the answers. The standard presentation of a carcass is defined as the body of a slaughtered pig, bled and eviscerated, without tongue, bristles, hooves, genital organs, flare fat, kidneys and diaphragm. Nevertheless, the standard presentation includes uncertainty related to the machinery and the slaughterhouse workers carrying out the slaughtering and preparation work. For example, the bristles are removed with a combination of scalding with hot water or steam followed by mechanical scraping, a process, which is known to differ from place to place. A very effective process can easily remove one mm of the skin surface compared to a less effective process, and as the area of a carcass surface is about one square meter, the influence on the weight is about one kilogram corresponding to about 0.5 LMP. Table 1 includes the total uncertainty based on an evaluation of the most important sources of error.

In a similar way the errors sources doing the dissection using a team of butchers are evaluated (Nissen *et al*. 2006) and included in table 1.

The factors influencing the CT data depend on the how the information is extracted (Olsen *et al* (2007). The resolution of images and the type of reconstruction algorithm, the specification of scanning parameters and the analysis of images are just some of the factors, which should be standardized to some extent. The results concern only an evaluation of methods based on segmentation analysis resulting in a number of voxels corresponding to tissue types, primarily meat, fat and bone. As an example, variation between scanners is estimated in a small study. Nine hams are scanned in three scanners within a short interval of time. The weight of each ham is estimated using the density estimates from a previous experiment. The estimated weights are compared to the scale weight. The average differences were –25 gram, 34 gram and 107 gram. The maximal influence on LMP is estimated at 1 percentage unit resulting in a Type B variance estimate equal 0.08 LMP. The final assessment is shown in table 1.

Table 1. Summary of uncertainty related to LMP working standard

	Uncertainty	Repeatability std.dev.
Standard carcass presentation	0.68 LMP	?
Knife dissection, EU reference standards	1.54 LMP	0.51 LMP
Computed tomography, without standardization	0.94 LMP	0.22 LMP

The scientific conclusions

Standard carcass presentation

No matter which reference method we use, we cannot avoid the uncertainty related to carcass presentation. In some extent the uncertainty can be reduced, but regional slaughter traditions will still be present and influence the reproducibility.

Manual dissection

The reference standards based on knife dissection are not ideal with respect to uncertainty. Even though an acceptable repeatability can be obtained by reducing the most influential factor, which is separating the carcass in specified parts, the differences between teams in the member states i.e. reproducibility is difficult to monitor.

CT based dissection

At the moment the only realistic alternative to manual dissection is the use of methods based on CT. Today, several CT methods have been used and no matter the type of method the agreement between LMP obtained with CT, and LMP obtained with knife dissection will be acceptable. This does not mean that all methods are equally good from a metrological point of view.

Summary

The outcome from European collaboration the last decade is a reduction of the uncertainty related to the manual handling, but primarily the repeatability part. The variability is hard to reduce – and monitor - because it primarily is related to different working conditions and methods at slaughterhouses throughout Europe.

There is potential to make a precise reference standard based on CT. However, more experience is needed. In particular, there is a need to determine how differences between scanners (brands and types (spiral or single slice images) and measuring protocols) can be managed. Effective and reliable reference materials (phantoms) might solve some of the problems. However, the type of image analysis also needs to be managed, i.e. how the data is modelled, the type of software and type of algorithms.

The next steps

The future work addressing the topic of obtaining an instrumental reference method based on CT independent of manual dissection, can be condensed into two items:

- Establishing a protocol for scanner parameters, which allows comparison of results from different brands of scanners. The work includes the development of phantoms and reference material.
- Developing a common method and algorithm to analyse data without any use of results from manual dissection.

Acknowledgement

The results have mainly been obtained from research and development financed by the Danish Pig Levy Fund and the Danish Government. The authors are very grateful for the collaboration with Danish Technological University, without that some of the basic research had not been possible. The authors also thank the technicians running the practical work, often at inconvenient hours of the day.

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Impact of pig population (light or heavy) on computed tomography (CT) and dissection relationship for lean meat percentage measurement

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Value for industry

- According to the EU regulation (EC n°1249/2008), all pig carcasses must be classified according to an approved classification method. The approval of a classification method consists in a calibration trial against a lean meat percentage (LMP) measurement reference method.
- The latest version of the EU regulation considers three different LMP measurement methods: total dissection of the left half carcass, partial four joint EU dissection carried out according to the Walstra and Merkus method (Walstra and Merkus, 1996). Total LMP measurement with a computed tomography (CT) scanner may be used, provided that the CT method of choice has been shown by the user to be equivalent to total dissection.
- The objective of this work is to achieve a new step in the definition of the terms of use of the CT in a calibration trial.

Background

Today, using CT as reference method for LMP measurement in a calibration trial is extremely complicated. Indeed the regulation does not specify the terms required to prove the equivalence between CT measurement and dissection. So these terms are discussed by the European experts in Brussels. The main requirement should be a good accuracy of the CT method involved. It must give results as close as possible as dissection results. Different EU countries research teams have already compared CT measurement (with different methods) and dissection (total or partial). According to all of these studies, the correlation between dissection and CT method is very high (r around 0.9) and the RMSEP in a case where the CT predicts the dissection is quite low (RMSEP around 0.5-1%) (Christensen and Boggard, 2005; Judas *et al.*, 2006; Romvari *et al.*, 2006; Font-i-Furnols *et al.*, 2009; Daumas and Monziols, 2011).

These teams have proved that the methods they have used has an equivalent precision to that of dissection (correlation and RMSEP). However, despite this, it appears that the EU experts consider that an alternative criteria must be met before CT can be accepted as a replacement for dissection. This criteria is the validity of the method on any pig population. So if dissection is assumed to be independent on the pig population studied, CT is not. In the end, even if a CT method has been proven to give extremely close results to the dissection ones on a previous trial, using it in a calibration trial requires at the expert demand additional dissections to show that the method is still valuable.

Why work is needed?

The main objective of this work is to demonstrate that a CT method is, as the dissection is assumed to be, independent of the sample. Such a study is needed to completely validate the use of CT as a reference method for the measurement of body composition as a replacement for dissection.

Methods used

This study was carried out during a calibration trial designed for the approval of grading methods in Italy. The Italian pig population was a unique occasion to achieve such a study. Indeed the Italian pig population is composed by 90% of “heavy pigs” and 10% of “light pigs”.

In the Italian Protected Designation of Origin (PDO) production of heavy pigs, different male breeds are used as stud boars: Large White or LW boar (11%); Landrace or L boar (2.4%); cross-breed 19.2%); hybrid boar (67.4%). Heavy pigs are slaughtered at 9 month age and about 160 kg live weight (+/ - 10%). Carcass weight ranged from 115kg to more than 150 kg. "Light" pigs are mainly Pietrain pigs slaughtered at about 120kg-130kg live weight with a carcass weight from 95kg to 110 kg. We can consider in this scheme that "light" pigs are closed to the classical European pig when the heavy pigs are typical Italian pigs.

100 carcasses were used in the study,25 heavy female pigs, 25 heavy castrated male pigs, 25 light female pigs and 25 light castrated male pigs . After 24h of chilling, the left half carcasses were prepared, cut into the four main joint (ham, shoulder, loin and belly) and dissected according to the procedure of Walstra and Merkus (Wastra and Merkus, 1996). Total weight and dissected muscle weight for each joint were recorded. And the reference LMP was calculated. We will call it the LMPEU.

Carcase size in heavy pigs was too large for the scanning zone of the CT, so all of the right half carcasses were cut according to the typical Italian cut into 5 joints, completely different from the EU joints and corresponding to the entire carcass minus the cheek.

All of these five joints were weighed, placed over a Styrofoam radio transparent support and analyzed by CT. The CT scanner used was a Siemens emotion duo (Siemens, Erlangen, Germany). The protocol of image acquisition had the following parameters : tube voltage 130 KV, tube current 40 mAs, FOV 500 mm x 500 mm, matrix 512 x 512, slice thickness 3 mm, spiral mode and reconstruction filter B30s (soft tissues).

Image analysis was performed automatically using software developed in C# (Monziols *et al.*, 2013) as described elsewhere (Daumas and Monziols, 2011). The examination table was removed from the image by an automatic ROI (region of interest) selection algorithm. The segmentation of the muscle voxels on the images was performed by a simple threshold between 0 and 120 Hounsfield unit (HU). The muscle weight of the joint was measured by multiplying the number of thresholded voxels by the voxel size (0.98x0.98x3) and a density fixed at 1.04 (ICRU, egpg).

A CT measure of lean meat percentage (LMPCT) was then calculated by dividing the sum of the muscle weights of the 5 joints measured by CT, by the sum of the total 5 joints weights. Statistical analysis was performed with the ANOVA and the generalized linear model procedures of R 2.14.1 (R core development team, 2008).

Results obtained

The table 1 shows the results of the half carcass weight, dissected LMP of the left half carcass and LMPct of the right half carcass for each population:

	heavy pigs		**light pigs**		**population effect**
	mean	Std	Mean	std	
left side weight (kg)	69,9	5,4	47,2	3,4	***
LMPEU	53,8	3,6	60,6	2,9	***
LMPCT	57,7	3,7	65,9	3,04	***

Table 1. Populations left half carcasses weights, LMPCT and LMPEU.

This table shows that there is a big difference between the two pig populations, the left half carcass is heavier, and both LMP measures shows that heavy pigs are largely fatter than light pigs. Furthermore we can see an important difference for both types between the two LMP measurements.

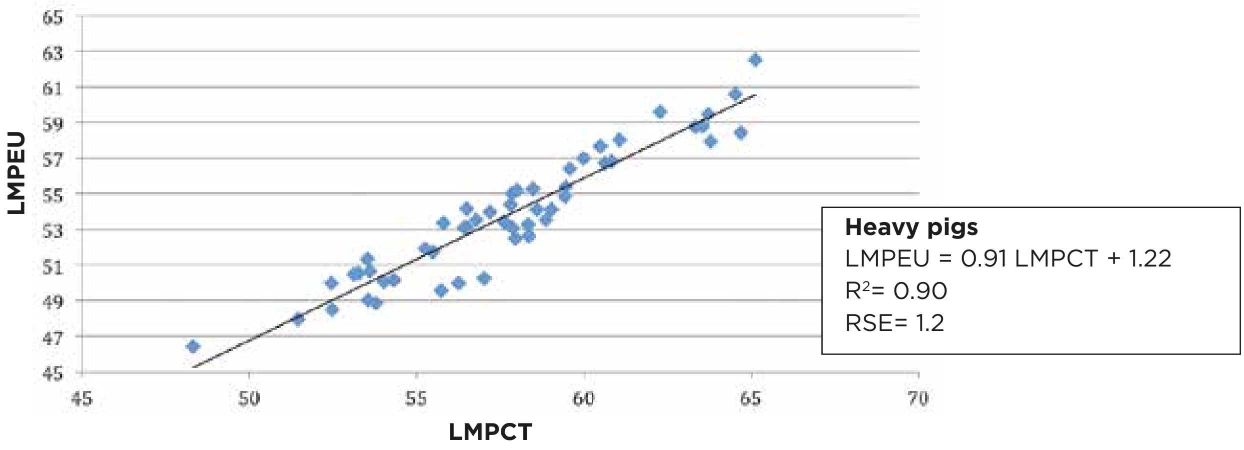
The main objective was to compare the relationship between LMPEU and LMPCT, so figures 1 and 2 present the results of a LMPEU prediction by LMPCT for each population.

Figures 1 and **2** show a high correlation between LMPCT and LMPEU. Furthermore residual standard errors around 1 are acceptable.

The main question was if there is a difference between these two relationships. In order to answer the question we pooled all data and use a different regression model in adding the population type and used the following model: LMPEU ~ LMPCT * population type .

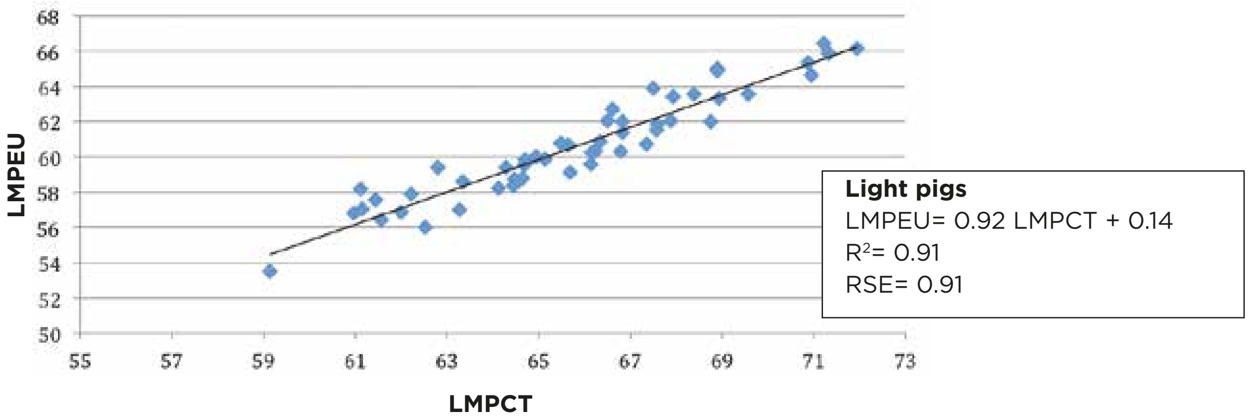
The **figure 3** shows the result of this model.

Figure 1. Relationship between LMPCT and LMPEU for heavy pigs.



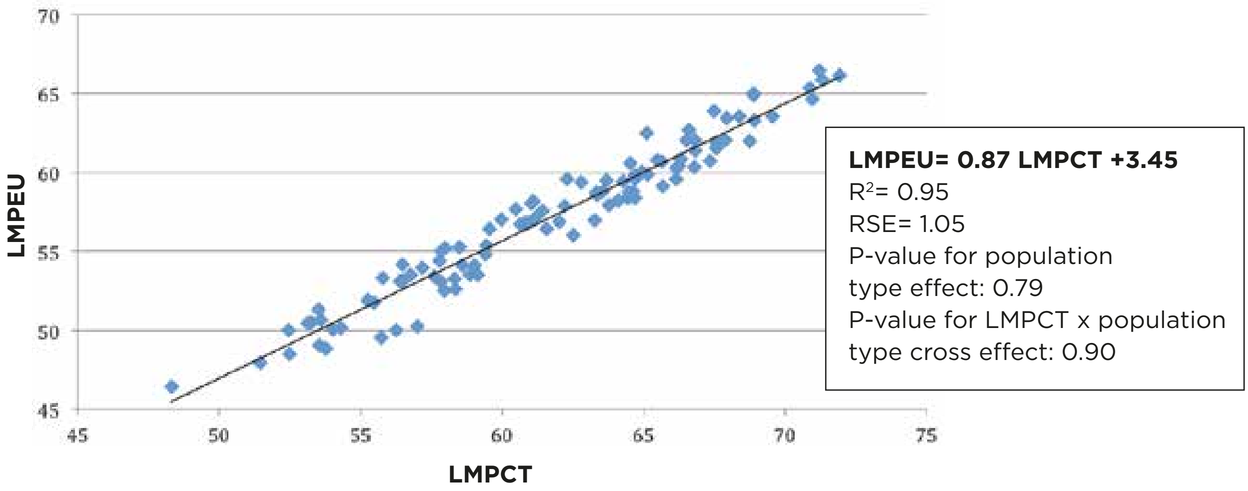
Heavy pigs
LMPEU = 0.91 LMPCT + 1.22
R^2= 0.90
RSE= 1.2

Figure 2. Relationship between LMPCT and LMPEU for light pigs.



Light pigs
LMPEU= 0.92 LMPCT + 0.14
R^2= 0.91
RSE= 0.91

Figure 3. Relationship between LMPCT and LMPEU for heavy and light pigs.



LMPEU= 0.87 LMPCT +3.45
R^2= 0.95
RSE= 1.05
P-value for population type effect: 0.79
P-value for LMPCT x population type cross effect: 0.90

The **figure 3** shows that the relationship between LMPEU and LMPCT is important for the whole population in this study, furthermore the residual standard error is still acceptable (very close to 1). However, the main result is that neither the effect of population type (light or heavy) nor the cross effect between LMPCT and population are significant, with two extremely high (0.79 and 0.90) p-values. This shows that the pig type does not contribute information to the model so the relationship between CT and dissection in this study is independent of the type of carcasses.

Scientific conclusions

The main conclusion of this study is that the relationship between a LMP measured by dissection or by CT in this study is clearly independent of the carcass size. Indeed in this study the two pig populations are considerably different with respect to carcass size. Heavy pigs half carcasses were 20 kg heavier than light pigs ones and their LMPEU was 7 points less than the light pig LMPEU. The Italian PDO pigs are very extreme and the difference between the two populations in this study may be unique in Europe. Nevertheless, the results show that in this study the type of pig has no effect on the relationship between LMPCT and LMPEU. Hence, since the relationship between CT and dissection appears to be similar in two extremely divergent populations of pigs, the results of the study suggest that calibration of a CT methodology does not need to be carried out in more than one population of animal.

The other conclusion of the study is that the relationship between an LMPCT method and the reference dissected LMPEU method is heavily dependent on the entities scanned and dissected. In a previous study (Daumas and Monziols, 2011), we showed a higher correlation (R^2=0.98) and a better RSD (RSD =0.54) than the results obtained in this study (R^2=0.95 and RSD = 1.04). But in the previous study, the same four main joints used in the dissection trial after EU cutting were scanned. In the present study the right side was scanned and the left side was dissected, and the dissection was made on EU joints whereas the CT scan was carried out on an Italian cut.
In both studies, the same CT machine with the same image acquisition parameters and the same image analysis method was used. So it may be reasonable to think that the differences between these studies come from the difference of entities scanned and dissected in the present work.
This result shows that it is really important for the comparison between CT measurement and dissection to perform both measurements on the same entity: joints or whole half-carcass.
For the LMP reference measurement, the EU regulation allows only the use of the CT method on entire half carcass, whereas the measurement by dissection can be done on half carcass or four main joints. In order to achieve parity for countries that want to continue with the current four main joints measurement methodology, and those who wish to use CT methods, it would be beneficial if the EU regulation also allowed LMP measurement by CT on the four main joints.

The next steps

This study is a very important step for the acceptance of CT based methods as a reference body composition measurement method.
It clearly shows that the measurement is independent of the animal population studied and that the accuracy of the relationship between CT and dissection measurements is more dependent on the entities compared : four main joints or whole half carcass.

For the use of CT methods for pig grading calibration trials, a change in the EU regulation would be needed to allow the use of CT as a reference method. To achieve this, a comparison trial against dissection is necessary, comprising the comparison of half-carcass or four joints by both methods.

For future steps, this work shows that a body composition measurement by CT method is not dependant of the type of pigs. This is a quality required for a reference method. Indeed, a unique reference CT method for body composition independent of the dissection must appear in a near future.

Acknowledgements

The authors want to acknowledge the agricultural Italian ministry, the Emilia Romagna Region and the FAIM COST for financial support of this study.

The authors want also acknowledge the Italcarni slaughterhouse for having hosted the entire study in their factory, and the Italcarni dissection team for their precise work.

In the end, the authors wants to say an important thank you to Enrica Gorlani, Andrea Bertolini , Michele Comellini, Eric Gault, Alain Le Roux and Arnaud Bozec for their technical support.

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Inter-laboratory comparison of medical computed tomography (CT) scanners for industrial applications in the slaughterhouses.

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Value for Industry

- Using computed tomography (CT) in the calibration of online grading equipment has been demonstrated to be beneficial over the last years by several institutions using medical CT scanners. The difference in makes and models calls for a standardized (and calibrated) method to be able to quantify differences in CT performance. The presented Round Robin scheme has demonstrated its potential as such a method.
- The benefit of the phantom set is that it provides a convenient way of comparing volume determination between different CT scanners. The suggested phantoms are mimicking important carcass features, conventionally recognized to be challenging to medical CT scanners.
- The web based classification software PigClassWeb has been demonstrated to be a convenient way of handling and comparing CT data in a transparent way, across regions and over time.
- The phantom set may be used to compare regional differences in analyzing software.

Background

For quite some years it has been suggested that CT might be used as a reference for determination of the lean meat content of pig carcasses (Dobrowolski *et al*., (2004); Romvari *et al*., (2006), eventually substituting or supplementing the EU reference based on manual dissection with a knife. One precondition for this suggestion is that the CT scanner is constructed to assess attenuation in biological tissue on an objective scale termed the Hounsfield Scale (Christensen *et al*., (2008); Olsen *et al*., (2007).The scale is referenced with a two point calibration to the attenuation in water and in air. Most tissues have attenuation values close to water, i.e. close to one of the calibration points. The CT scanner hardware and software is calibrated to a high accuracy with respect to volume as one major application of medical scanners is determination of tissue volumes. The settings of scanner parameters are optimized for a specific diagnostic purpose thus optimizing the medical evaluation of tissue parameters of relevance. The optimization is a highly proprietary setting of scanner parameters, obscured for the user.

Why work is needed

The obscurity calls for use of an inter-calibration procedure to support transparency of reference data produced on different makes of scanners. Round-Robin testing is widely known from international standardization work e.g. on acoustics (sound pressure and acceleration) and geometrical coordinate measuring systems. The Round-Robin procedure is based on circulation of a stable work piece, a phantom, between laboratories thus making measurements performed at different locations directly comparable. As the claim for stability of the phantom is a major challenge in measurement of lean meat content (LMC), the substitution of the relevant tissues with stable materials is necessary. One other feature of the CT_{LMC} phantom is the ability to reveal instrumental differences, not only between scanner makes and models but also between analyzing software.

The method used

To handle the challenge of measuring partial volumes of heterogeneous objects we have included a volume measurement of the individual materials composing each phantom in the set of seven. The pycnometer method, based on water displacement, is preferred to determine the reference volume of all materials in the set of phantoms before assembly.

The phantom set is designed to cover the range of lean meat content from 52% to 70% by changing the ratio of the polymers representing fat and meat tissues respectively.

The polyethylene (PE) polymer was chosen to represent fatty tissue and the polymethyl methacrylate (PMMA) polymer was to represent lean meat. To be able show scanner performance on pig carcasses, the design of the phantoms simulates a piece of middle, including back and belly parts. The rectangular shape of a pig middle is known to be a challenge for the reconstruction algorithms of medical scanners, as they are optimized for cylindrical objects like a head, torso or the extremities of human. An exploded view of one of the phantoms is shown in Figure 1.

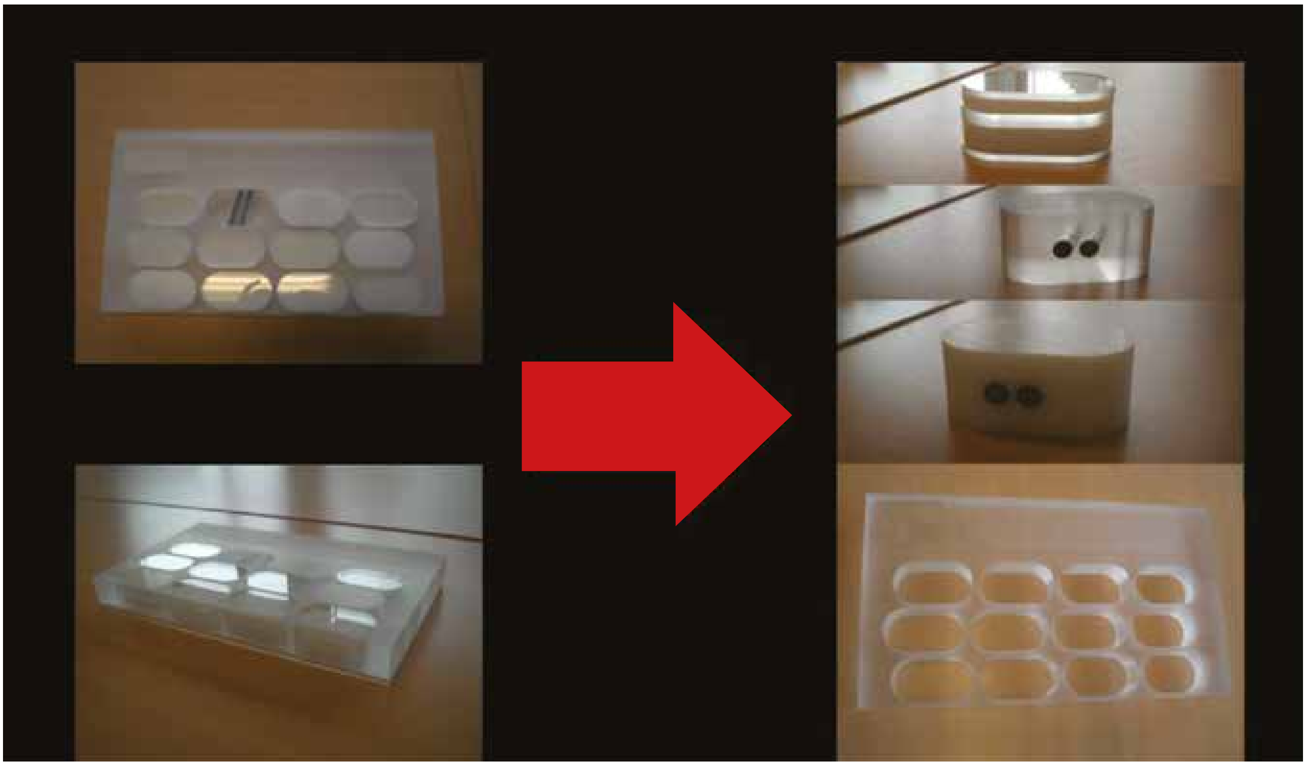

Figure 1. The belly phantom. One of the members of the belly phantom set assembled (left) and dismantled (right). The different members of PE (white), PVC (black) and PMMA (clear) are easily distinguishable. Before assembly the volume determination of the three polymers is made using a pycnometer.

As a feasibility study of the Round-Robin procedure two selected samples from the phantom set are measured by four different institutions over a period of approximately 12 months. The results are compared to the reference values from the pycnometer method.

Generally speaking, one important feature of a phantom is its ability to reveal relevant parameters of the instrumental method. In Figure 2 an example of measurement of three different phantoms from the same scanner are displayed; one standard QCT phantom developed for medical purposes, one of the phantoms in presented phantom set and one phantom developed for testing our online CT scanner. The phantoms demonstrate different scanner features. The medical phantom is a scale calibration phantom with six different materials of calibrated Hounsfield readings but very limited challenges to the geometrical features in the reconstruction, the belly phantom shows the complete link of volume analysis, whereas the middle phantom challenges the scanner in dynamic range, automated feature extraction (minimum thickness) and geometrical accuracy.

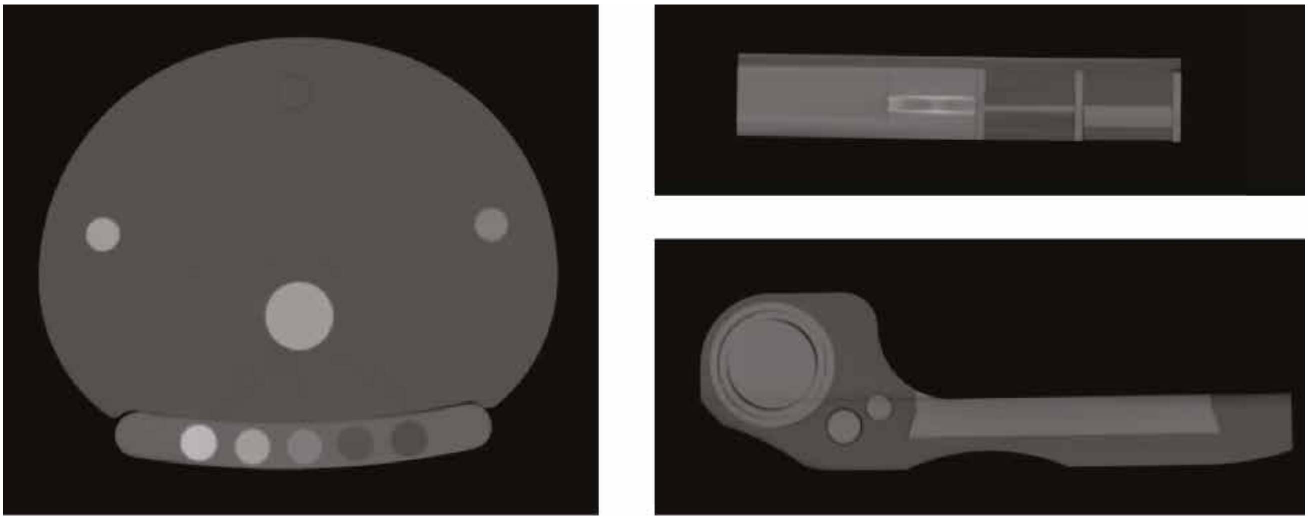

Figure 2. Three examples of CT phantoms, measured on the same scanner using the same look-up-table: Left is a medical QCT phantom containing six different attenuating polymers each with a different Hounsfield reading. The phantom is only a moderate geometrical challenge to CT scanners and contains no volume information. Right (top) is a scanning of a member of the belly phantom set, stressing the geometrical performance due to the elongated shape. The phantom is volume calibrated before assembly. Right (bottom) is a middle phantom designed for use in development of an online CT scanner. The middle phantom is geometrical calibrated (thickness).

The results obtained

We have demonstrated the potential of automated, web based image analysis by designing the Pig-ClassWeb home page (Christensen *et al*., (2010). The automated workflow has been beneficial for carcass scanning, facilitating the access to data in a very standardized format so reducing the risk of operator induced errors. A web based tool might be developed to handle phantom CT measurements, data analysis and documentation. The tool may ease the comparison of scanner performance to analysis stability over time.

Round-Robin results

As a preliminary demonstration of the Round-Robin procedure two samples are circulated between four Europeans CT institutions. The preferred protocol used for carcass scanning by the institution must be used for scanning of the phantoms. The protocol parameters are summarized in table 1. The parameters are not freely selectable but must be chosen from a predefined range available on the scanner console. The parameter setting is known to influence the results of the volume determination.

Table 1. Setting of two important scanning parameters in the different scanners included in the Round Robin scheme.

	ID 1	ID 2	ID 3	ID 4	ID 5	ID 6	ID 7
Slice thickness [mm]	10	1	2	3	10	1	10
Focal Spot size [mm]	0.7	1.2	0.95	0.95	NA	1.2	0.7

Automated image analysis of the CT scanned carcasses may be done with a large number of methods and strategies. The belly phantom has been used to compare three different methods: A Full-Width-Half-Maximum (FWHM), A contextual Markov Random Field method (Owen-Hjort-Mohn (OHM) (Vester-Christensen *et al.*,(2009) and a quite basic Threshold method. Two of the phantoms are measured in triplicate and the lean meat percentage as determined by CT (LMPCT) are determined using the three classifying methods. The results are seen in Fig. 3.

The volume of PE (fat) and PMMA (lean meat) determined by the three methods compared to the pycnometer reference are given (in ml) in table 2 below. The deviations to the reference are much higher than the uncertainty of the reference method itself being in the 5 to 10 ml range.

The agreement between the quite complex contextual and the basic threshold based are striking whereas the FWHM deviate significantly from the two other methods. These findings are supported by previous measurements on pig bellies (Christensen *et al.*, (2008).

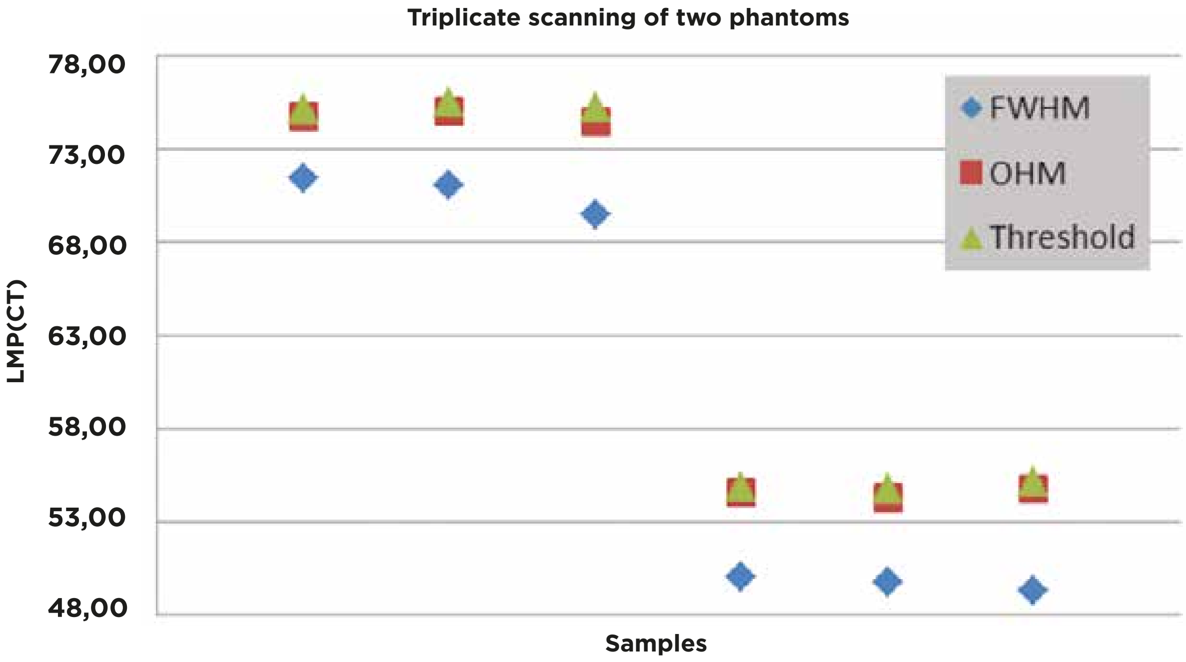

Figure 3. LMPCT determined in triplicate of two of the belly phantom members, one lean and one fat. The result of the six identical scanning protocols is analyzed by different classification software. Full-Width-Half-Maximum, Owen-Hjort-Mohn and Threshold (Vester-Christensen *et al.*, (2009) demonstrate variation in performance on the phantoms.

Table 2. Volume measurement of the two principal polymers of the belly phantom members 1 and 7, respectively. The pychnometer reference in ml is directly comparable to the volumes determined by the three classifying methods. The FWHM seems to be outperformed by the two reminder methods.

Phantom	Material	Pycnometer	FWHM	OHM	Threshold
Phantom 1	PMMA (Lean meat)	3819.9	3880.0	4089.7	4153.7
	PE (fat)	1342.5	1741.6	1487.8	1467.8
Phantom 7	PMMA (Lean meat)	3026.5	2787.0	3037.0	3084.2
	PE (fat)	2135.2	2832.0	2550.0	2534.8

The correlation between the four volume reference readings and the corresponding CT volumes assume 0.934, 0.987 and 0.990 respectively, again leaving the OHM and the Threshold methods superior to the FWHM approach. The Round-Robin results on volume determination of PE and PMMA are given in Fig. 4 below

The deviation from the reference volume is in the 5% to 10% range for all measurement sessions but ID 7 being significantly higher.

The results from the Round-Robin scheme described above include the entire chain of accuracy with contributions from scanner hardware calibration, scanning protocol and image analysis from each of the participants. These preliminary results can be taken as a demonstration of the potential of a Round-Robin scheme for quantification of the accuracy of CT scanning but also as a way of making a detailed analysis of the individual contributions from hardware and software.

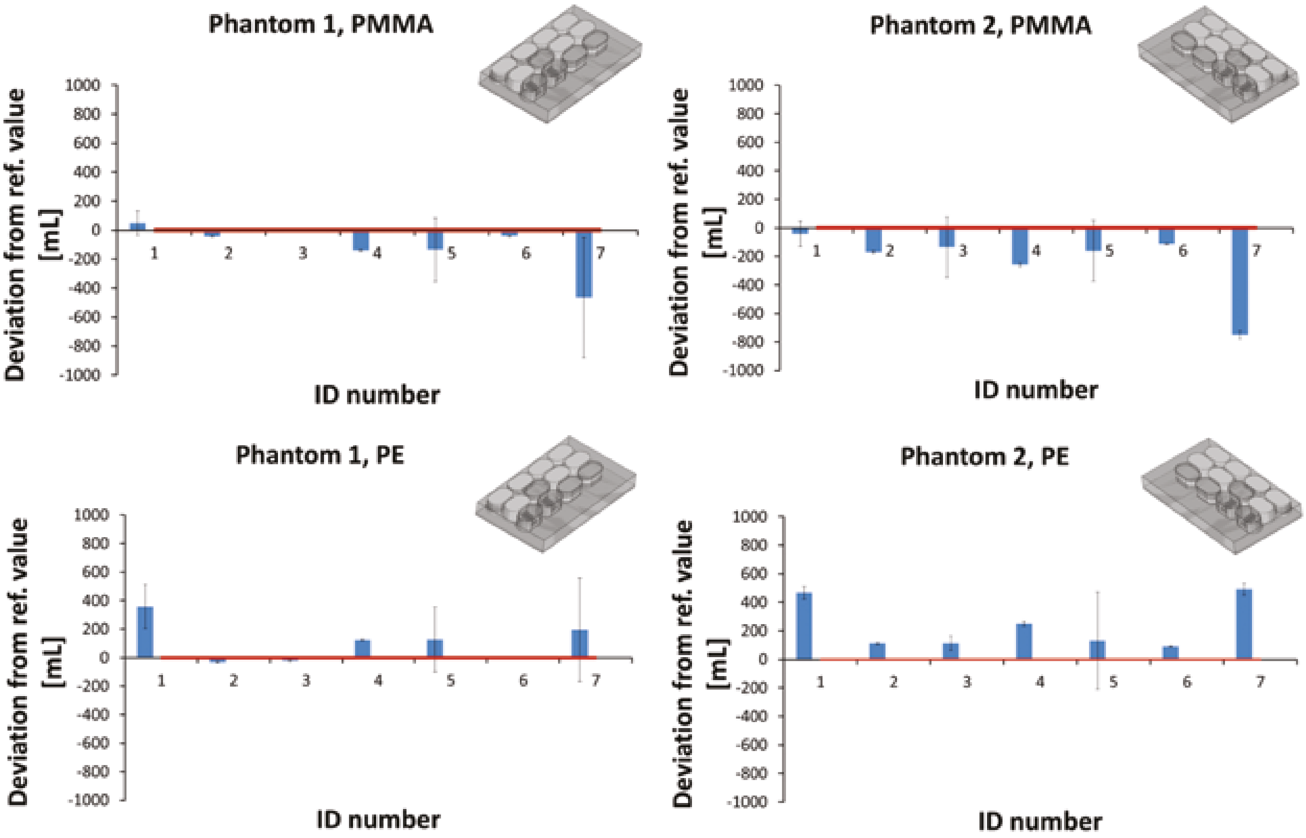

Figure 4. Volume performance of the seven participants in the preliminary Round Robin scheme. Comparing to the reference volumes in Table 2, a deviation in the range of 5% to 10% is demonstrated.

The scientific conclusions

Pycnometer

The pycnometer measurements define the reference values for the volume of the individual polymers of the phantoms. The reference values are made before assembly of the phantoms. After assembly the phantoms are vacuum packed to keep the different tissue compartments forming a stable rigid construction. The method has demonstrated a high level of feasibility during the 12 month test period

The accuracy of the reference measurement by far outperforms the CT determination of the volume, suggesting that the pycnometer method is a suitable reference for volume in this context.

Software comparison

With respect to the three image analysis strategies reported above the FWHM method seems to deviate significantly from the OHM and Threshold methods. The same conclusion may be made for the individual volume determination. Compared with a pycnometer reference, the FHWM method seems to be out performed by the OHM and the Threshold methods.

Hardware comparison

The stability of the phantom ensures a long term analysis of different CT hardware is possible and a standardized software analysis could facilitate such analysis and documentation.

Protocol comparison

The main preliminary results indicate the superior performance in volume determination of the phantom materials of the smaller voxel sizes, slice thickness of 1 and 2 mm respectively. It still has to be confirmed that the slice thickness alone is due to the appearing better volume determination.

Round-Robin procedure

The Round-Robin scheme has been demonstrated to be a suitable way of monitoring the performance of CT scanners over Europe. The duration of a scanning session of one week seems to be acceptable and applicable for the participating institutions. This includes all practical handling and scanning of the phantom set.

Summary

The feasibility of the suggested Round-Robin scheme to quantify differences in volume determination of polymer materials mimicking lean meat and fat has been demonstrated within a limited group of European CT institutions. Furthermore the aim has been to establish a procedure that may be conducted to verify any drift in instrumental volume determination and consequently in Lean Meat Content measured with Computer Tomography.

The next steps

The future work will be the inclusion of more institutions in a Round-Robin scheme on the complete 7 piece phantom set. This work should focus on separating the individual contributions to the differences observed. At a certain point the group of institutions might open a discussion with the European Commission concerning the benefits of including a Round-Robin scheme as a standard procedure in the European classification standardisation initiative.

Acknowledgement

The results have mainly been obtained from research and development financed by the Danish Pig Levy Fund and the Danish Government. The authors are very grateful for the collaboration with Danish Technological University, without that some of the basic research had not been possible.

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*Winner of the poster competition

Can computed tomography of live animals be a tool for scoring osteochondrosis in a pig breeding program?

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Value for industry

- *In vivo* assessment of osteochondrosis by use of computed tomography (CT) can enable phenotyping of live animals.
- A non-invasive method will potentially increase genetic gain of the trait by increasing the accuracy of the breeding values.

Background

Osteochondrosis can lead to lameness (Reiland, 1978). In large scale data collection, the condition is traditionally assessed in two ways: macroscopically inspection of the joint cartilage surface or by sectioning the joint in slabs and scoring the slabs (Grøndalen, 1974). Both methods are applied post mortem. From a breeding perspective assessing osteochondrosis (OC) *in vivo* would allow direct phenotypic assessment of the individual candidate for selection. A phenotypic assessment on the selection candidates has several advantages most of them will ultimately increase genetic gain if the trait of interest is included in a breeding goal.

Why work is needed

Genetic improvement for a trait often results in unplanned development in other traits (J. Conington *et al*., 2010). For osteochondrosis, a deterioration can be expected when young animals undergo selection for growth rate (Aasmundstad *et al*,. 2013). To minimize risk of unwanted changes, cost-effective *in vivo* assessment for measuring the phenotype on the breeding animal itself is beneficial.

The methods used

In the pig breeding program of Norsvin, all boars are subjected to CT scans (Kongsro, 2012). From the start in 2008 to current, this adds up to >17 000 boars scanned. After scanning, the images are compressed and stored on an image server that gives access for further processing of the images. At present, as described by Gjerlaug-Enger *et al.* (2012), carcass yield and meat percentage are two of the phenotypes calculated from the CT information at scanning.

The 2273 boars in the study were sedated by an intramuscular injection of Stresnil Vet® prior to scanning. The scanning procedure took about 5 minutes for each animal and yielded output images for each 1.25 mm with a pixel size of 0.93 mm^2. The scanner used was a General Electric 32 slice VTC LightSpeed. The high resolution (pixels*slice thickness) permits a 3D visualisation of the individual bones of the skeleton as shown in figure 1. The assessment for OC in the material studied was done by one trained assessor who used less than 5 minutes to assess all eight anatomical locations on one animal. The scale of assessment is shown in figure 2, and is based on the assessment scale developed by Grøndalen and Lium (Ytrehus *et al.,* 2004) for scoring joint sectioned into slabs. The method is further described by Aasmundstad *et al.* (2013). The OsiriX software (Rosset *et al.*, 2004) was used in the assessment situation and a threshold window level of 400 HU and a window with of 400 HU were set to get the best contrast/detail level. This window level was also suggested by Ohlerth and Scarf in 2007. A pilot study in the project reveals that the positive predictive value of examination by CT is 100% when compared to histological evaluation (Olstad *et al*., unpublished). Figure 3 shows a typical scoring scenario; the same joint is visible in 2D simultaneously for three planes; axial, coronal and sagittal. For the preliminary statistical handling of the phenotypes the SAS/STAT® software were utilized, while estimates of genetic variance components are output from the DMU 6.7/AI-REML software package (Madsen and Jensen, 2008).

Figure 1. First image: 3D model based on CT posterior end of a boar. The boar shows signs of OC on femur medial condyle. Second image: 3D model based on CT of the left femur of a boar. OC lesions can be seen both on the medial and lateral condyle.

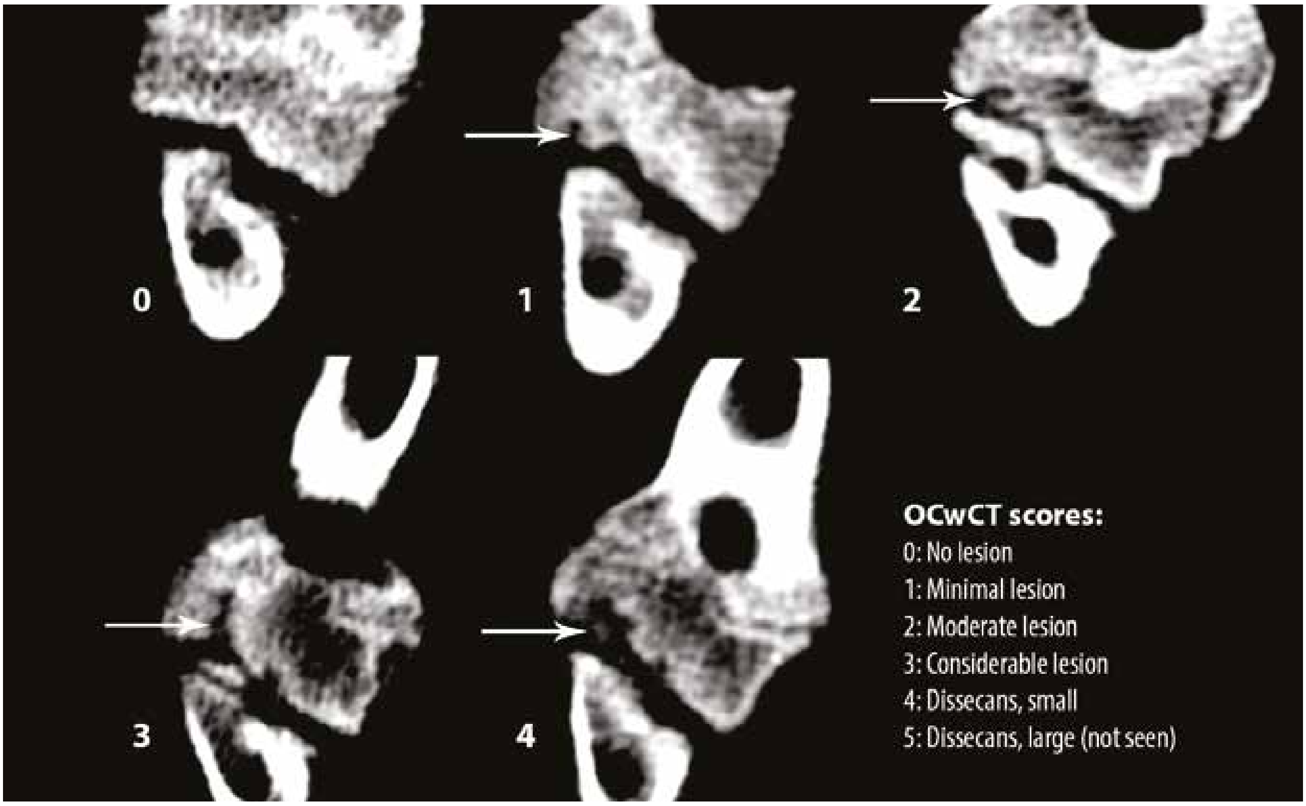

Figure 2. Scoring of osteochondrosis in the distal humerus and femur in pigs with computed tomography. 0 denotes no lesion, 1 denotes minimal lesion, 2 denotes moderate lesion, 3 denotes considerable lesion and 4 denotes small dissecans. 5 denotes large dissecans, not seen in the material. All images are from the right medial condyle of humerus.

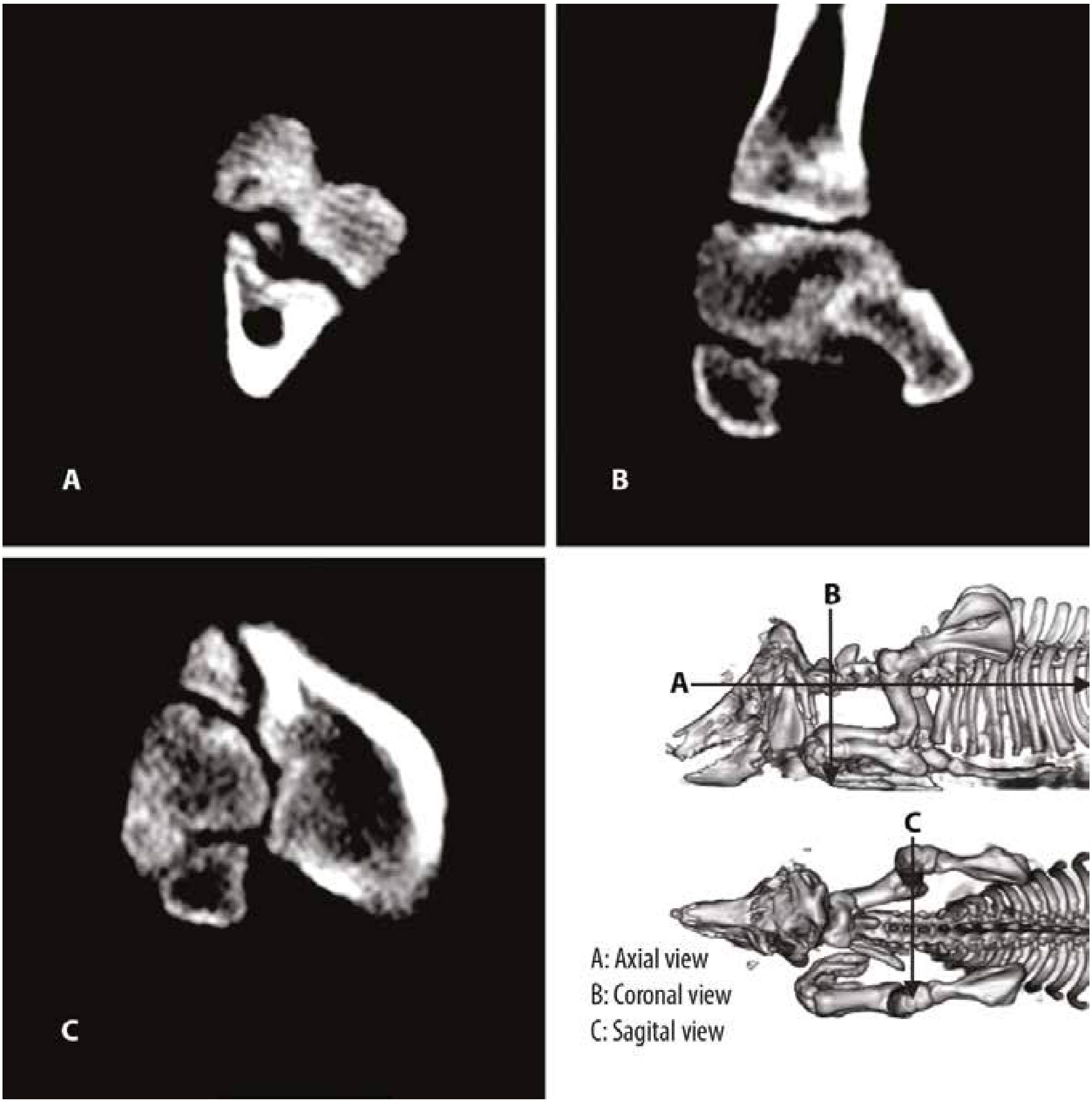

Figure 3. CT images showing information available in a scoring situation for humerus. Lower left quarter of figure shows a 3D reconstruction of the skeleton with arrows indicating the assessment planes. Upper case letters corresponds to letters in the three quarters of the figure.

The results obtained

Data from the 2273 boars showed that lesions can be found at all sites, but with varying frequencies (table 1). About 15% of the boars are completely free from lesions. Of the eight anatomical positions assessed, the lowest frequencies were found on lateral side of the humeral condyles were ~97% of the animals were found to be free from lesions. On the contrary, only ~39% of the animals were devoid of lesions on the medial condyles of femur. Evidences for loose bone fragments (osteochondrosis dissecans (OCD) in the population at study were scarce, only 5 out of 2273 boars (0.22%) had signs of OCD.

Table 1. Percentage, means and s.d of pigs appearing in each osteochondrosis lesion score category and anatomical location.

Position	HRM	HRL	HLM	HLL	FRM	FRL	FLM	FLL
Lesion score								
0	75.1	97.49	75.05	97.93	38.58	81.08	38.8	80.6
1	9.11	1.28	8.93	1.19	51.3	15.79	50.86	16.76
2	10.29	0.97	10.34	0.75	9.59	2.51	9.99	2.11
3	5.46	0.22	5.46	0.13	0.53	0.57	0.35	0.53
4	0.04	0.04	0.22	0	0	0.04	0	0
Mean	0.46	0.04	0.47	0.03	0.72	0.23	0.72	0.23
s.d	0.89	0.28	0.9	0.23	0.65	0.52	0.65	0.5

Anatomical abbrevations: First letter position H=Humerus, F=Femur. Second letter position: R=right, L=left. Third letter position: M=medial, L=lateral. For example, HRM=Humerus right medial. All scores are based on computed tomography. Number of pigs assessed=2273.

Heritabilities for anatomical locations and the total score (OCT) can be found in table 2, as can also the genetic correlations amongst the traits. The heritabilities for the different anatomical positions varied with a highest heritability (h^2=0.28) found for the medial condyle of the right humerus.

The heritability of OCT was 0.29. Genetic correlations between the anatomical locations and OCT were overall favourable and medium to high.

Table 2. Heritabilities and genetic correlations for the individual locations and the sum of the locations. All estimates are from bivariate analysis. N.E indicates analysis that did not converge within the set criterion.

	OCT	HRM	HLM	FRM	FRL	FLM	FLL
OCT	0.29 (0.06)	0.92 (0.04)	0.96 (0.04)	0.65 (0.13)	0.62 (0.17)	0.63 (0.14)	0.56 (0.41)
HRM		0.28 (0.06)	0.99 (0.03)	0.34 (0.19)	0.37 (0.24)	0.17 (0.22)	0.06 (0.45)
HLM			0.27 (0.06)	0.33 (0.2)	0.34 (0.24)	0.35 (0.21)	0.39 (0.47)
FRM				0.12 (0.04)	0.22 (0.31)	0.89 (0.13)	-0.15 (0.6)
FRL					0.07 (0.03)	0.04 (0.37)	N.E
FLM						0.09 (0.04)	0.23 (0.57)
FLL							0.02 (0.03)

The scientific conclusions

- CT may be used as a tool for assessing osteochondrosis *in vivo.*
- The trait "sum score of osteochondrosis" (OCT) has a medium heritability.
- The genetic correlations between the eight individual locations investigated and OCT are high, suggesting that a selection for reduced OCT would reduce incidence at all locations.
- As the selection candidate is subjected to CT, a phenotype for the candidate itself will improve accuracy of the breeding value and hence potential of genetic gain.

The next steps

- Investigate possibility for semi-automatic phenotyping of OC.
- Develop a technique to distinguish the lesions subjected to repair from that of a loosening bone fragment.
- Implement OC as a selection criterion in a pig breeding goal.
- Use CT to study longitudinal development of OC lesions in growing pigs.
- Investigate phenotypic and genetic relationship between conformation traits and osteochondrosis.
- Investigate potential genetic link between OC and sow removal.

Acknowledgements

The study presented is a part of the "Robust pig" project funded by the Research Council of Norway (project no: 199598).

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Testes characteristics of entire and immunocastrated male pigs from 30 to 120 kg live weight measured *in vivo* with computed tomography

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Value for industry

- Computed Tomography (CT) is a non-invasive technique which can be used in live animals.
- CT images are represented by a large number of X-ray attenuation values (CT values) which are expressed in Hounsfield units (HU). Body tissues that have been scanned show different densities which permits to distinguish and quantify them.
- At present, surgical castration is used to prevent boar taint. The best alternative to prevent boar taint appears to be immunocastration; the vaccination against gonadotropin releasing hormone (GnRH).
- The use of CT scanning in live pigs can give an indication of the effectiveness of immunocastration.

Background

Computed Tomography (CT) represents one of the best ways to study the body composition of live pigs. CT takes images that are represented by a large number of X-ray attenuation values and shows different densities of the body tissues that have been scanned. Attenuation of CT values is expressed in Hounsfield units (HU). So, knowing these different densities allows lean, bone and fat tissues to be identified and quantified. A detailed description of the CT techniques is given by Allen and Leymaster (1985). Live pigs can be fully scanned and CT images can be visually and numerically analyzed. This technique allows different types of pigs to be scanned and their tissue development during growth to be followed.

Rearing entire males (EM) has benefits in terms of their high production efficiency and lean meat content; however, the risk of boar taint precludes the acceptability of boar meat in most countries. To prevent tainted meat reaching consumers, the present practice is to castrate male piglets early in life. This practice (surgical castration without anaesthesia) is questioned, since there are clear physiological changes and indications of pain suffering even at this early age (Prunier *et al.,* 2006). Due to obvious animal welfare issues, the European Union (EU) wants an alternative to surgical castration. The European Commission and representatives of European pig farmers, meat industry, traders, retailers and scientists have recently committed themselves to plan to voluntarily end surgical castration of pigs in Europe by January 1st, 2018 (European Declaration on alternatives to surgical castration of pigs; SANCO 2010). Immunocastration by vaccination against gonadotropin releasing hormone (GnRH). is a viable alternative to surgical castration. Vaccination has to be performed twice in an interval of at least 4 weeks And the mechanism of action is based on the immune system of the pig recognising and attacking the injected GnRH analogue peptide in a process called immunoneutralisation. With no GnRH, no luteinizing hormone (LH) and no follicle-stimulating hormone (FSH) are produced and, consequently, the Leydig and Sertolli cells that produce testosterone and sperm respectively become inactive. Hence, the loss of GnRH results in cessation of reproductive function.

Many authors have studied changes in testes weight due to immunocastration (Jaros *et al.,* 2005; Gispert *et al.,* 2010; Soede *et al.,* 2011; Kubale *et al.,* 2013).

Moreover, the study of histomorphologic properties of testes such as: the length of *tubuli seminiferi,* the height of *seminiferus epitelium*, the number of Leydig and Sertolli cell and their nucleus area, have been also reported (Caires *et al.,* 2008; Kubale *et al.,* 2013) and have shown significant differences between immunocastrated male pigs (IM) and EM in testes growth. But no study has so far reported changes in testes density.

The aim of this experiment was to evaluate testes density and volume in EM and IM from 30 to 120 kg live weight (LW) by analysing CT images.

Why the work is needed

Research has shown that testes size is not a reliable indicator of successful immunocastration i.e. of boar taint prevention (Frederiksen *et al.,* 2011), at least, not within recommended delay (4-6 weeks) between the second vaccination (V2) and slaughter (Kubale *et al.,* 2013). Bonneau (2010) demonstrated that shrinking of reproductive organs is proportional to the length of the interval between V2 and slaughter, and that seminal vesicle could serve as an indicator of successful immunocastration. No studies have so far related testes density of live pigs during the growing period, prior and after the first vaccination (V1) and V2. This information can be used to relate with testes size, effectiveness of vaccination and boar taint in further experiments.

The methods used

The experiment involved 24 pigs, 12 EM and 12 IM. The experimental period was from 30±2.0 kg to 120±3.9 kg LW. All the pigs were weighed weekly and fed *ad libitum* with a commercial diet.

Immunocastration vaccine Improvac® was injected twice, at 12 (V1) and 18 (V2) weeks of age. Live pigs were scanned at the target LW of 30, 70, 100 and 120 kg and also a subsample of 5 animals of each sexual type two weeks after V2 (at 83.3 3 2.3 kg LW). V2 coincided with the scanning at 70 kg target LW.

The scanning

The CT scanner used in the present study was a General Electric HiSpeed Zx/I, located at IRTA-Monells. Live pigs were scanned and prior to scanning they were fasted for at least 8 hours and weighed. Intramuscular sedation was used at all target LW and at 100kg and 120 kg an intravenous sedation was also applied. No animal died during the experiment. When the animals lost consciousness they were placed in a cradle, constrained by a set of belts, and moved into the CT unit (Figure 1). CT measurements were performed at 30, 70, 100 and 120 kg target LW. The procedures were approved by the Ethical Committee.

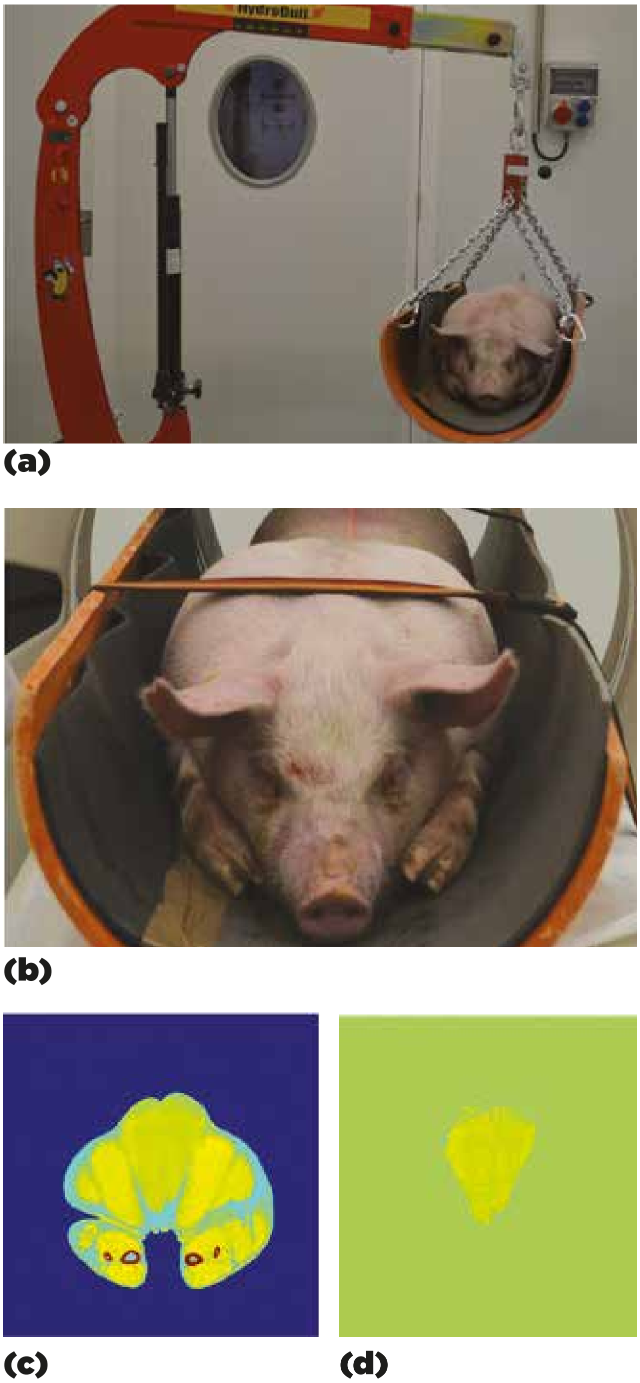


Figure 1. CT scanning of pigs: **(a)** lifting and moving pigs with a crane, **(b)** pig inside a PVC cradle ready to be scanned, **(c)** rear part section that includes the testes, the ham and the legs **(d)** testes image extracted from image on Fig. 1c.

Image analysis

To evaluate the testes, images from testes section were processed to obtain the volume and the density. CT image analysis was carried out with the help of Matlab® scripts, written inhouse, to obtain volume and average Hounsfield values (HU) of testes.

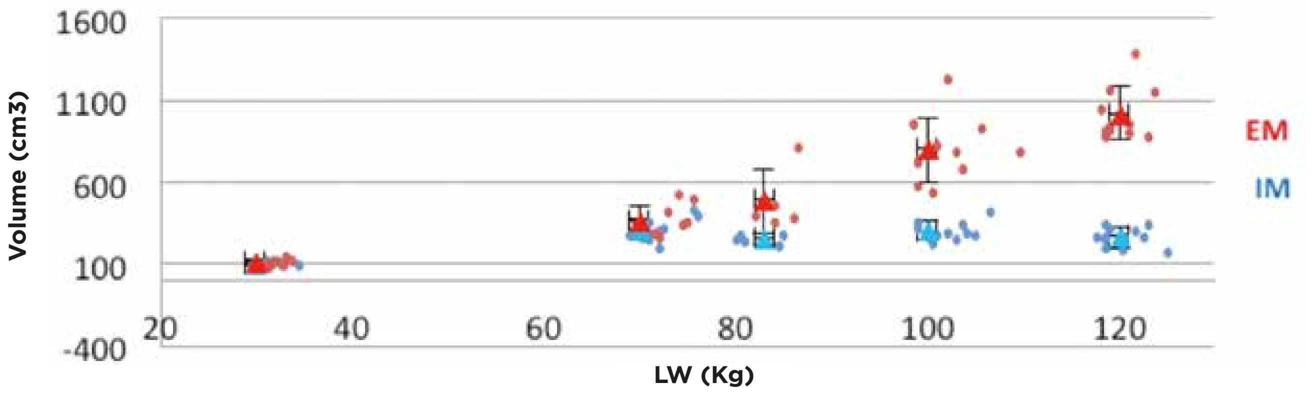
Statistical analysis

Statistical analysis was performed using SAS software (version 9.2., SAS Institute Inc, Cary, NC, USA, 2001). Mixed procedure was used to determine differences between treatments. The model included sexual type, target LW and their interaction as fixed effects.

The results obtained

Mixed procedure of SAS determined a significant effect of sexual type (P<0.0001), target LW (P<0.0001) and their interaction (P<0.0001). No differences in testes volume were observed between EM and IM at 30 and 70 kg LW. However, two weeks after V2 (at 83 kg), EM had bigger testes than IM (522.72 *vs.* 296.04 cm^3, P=0.0136, respectively). These differences became even more important (P≤0.0001) at 100kg (V2 + 3.5 weeks) and 120 kg (V2 + 6 weeks). Testes density analysed by averaged HU value showed that, at 30 kg and 70 kg LW, respectively, there were no differences between two sexual types, but this changed two weeks after V2, when the vaccination became effective. Results revealed that testes of IM were denser at 83 kg LW (P=0.003) and 100 kg (P=0.027), however, the differences became non significant (P=0.553) at 120 kg (Figure 2).

(a) **Volume of IM and EM testes**



(b) **HU's mean of IM and EM testes**

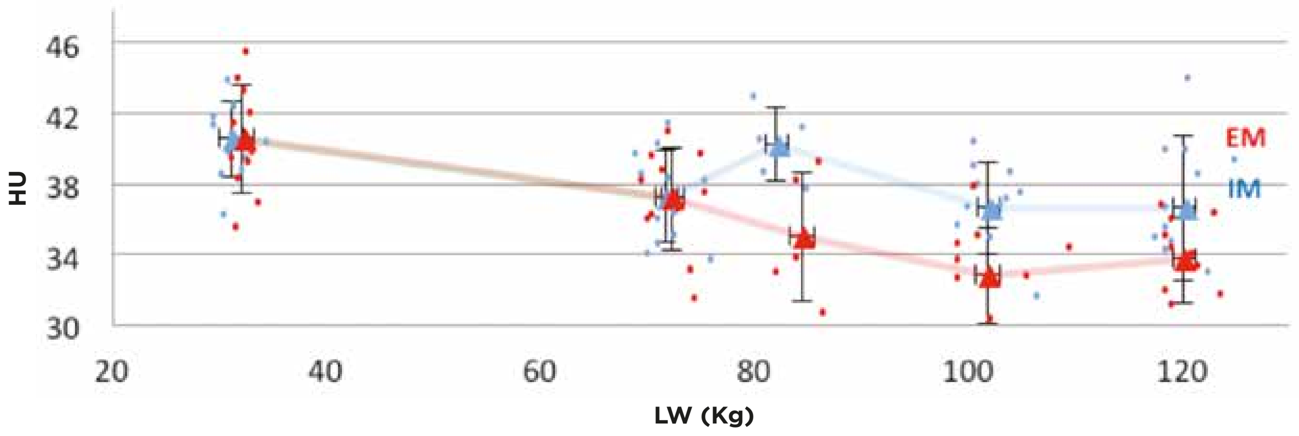


Figure 2. Volume (a) and density in Hounsfields (HU) values (b) of immunocastrated (IM) and entire (EM) male testes at each target live weight (LW).

The scientific conclusions

The majority of information about the biology associated with spermatogenesis has been gained with the use of rodent models and as a result, there remains a general lack of knowledge regarding testes development and sperm production in livestock. Further, due to the complexity involved, most research investigating testes is often restricted to *in vitro* studies. Although CT does not permit to study a tissue with microscopic precision, it can give an accurate estimation of tissue density during growth in individual animals. Density is the relation between mass and volume; in EM, the volume of the testes increased (P<0.05) with increasing body mass, whereas in IM, the volume of the testes increased (P<0.05) between 30 and 70 kg LW (until V2, when they were physiologically entire males). Thereafter testes volume in IM did not increase (P>0.05). After V2, at 83 kg and 100 kg LW, IM had denser testes than EM. Kubale *et al*. (2013) studied the reproductive organs and histologic parameters of IM pigs from 12-24 weeks of age. They reported that histomorphologic properties of testes were affected by immunocastration (post V2) with Leydig cells being progressively smaller, losing their polygonal shape, *tubuli seminiferi* were shrunken, empty spaces were visible inside tubules and a progressive reduction of epithelium height was noted. In agreement with our results, this suggests that all these histomorphologic changes could be responsible for the changes in density of IM testes.

More importantly, these findings (Kubale *et al.*, 2013) indicate that, probably, density would be a good indicator of the effectivness of the vaccination. In the present study, density differences (P<0.05) were clearly discernible between 70 kg and 83 kg LW, that means, two weeks after the second dose of the vaccine, in agreement with studies of Lealiifano *et al.* (2011) and Kubale *et al*. (2013) proving effectiveness of vaccination already 2 weeks post V2.

The next steps

More studies are needed to clarify biological activity/function of the testes in IM in relation to the tissue density of the testes in live animals. It would also be of interest to study any relationship between density changes and the concentration of boar taint compounds. Future experiments may provide insights for both agricultural and biomedical applications.

Acknowledgements

The present research study was supported by the Instituto Nacional Investigaciones Agrarias (Evaluación *in vivo* del crecimiento alométrico de los tejidos muscular y adiposo de los cerdos según la genética y el sexo mediante tomografía computerizada. Núm. Proyecto: RTA2010-00014-00-00). Scientific collaboration of M. Čandek-Potokar was financed by Slovenian agency of research (P4-0133).The authors wish to thank Albert Brun, Carles Francàs, Albert Rossell, Agustí Quintana, Alfons Varas, Albert Fonquerna and Carlos Millán for their invaluable technical assistance.

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Poster

Gemma Curtis

Carcass composition of Holstein bull calves determined using Spiral Computed Tomography

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Introduction

The U.K. dairy industry currently produces approximately 500,000 Holstein bull calves annually. These animals have previously been deemed as 'waste', unsuitable for use within the dairy and beef industries.

However recently, dedicated supply chains have been set up to reduce exports and make use of these animals by incorporating them into the beef and rosé veal industries[1].

Holstein calves intended for both the Dairy and Beef industries, traditionally reared on least cost principles, are often limit fed milk replacer (MR) and weaned onto solid feed by 6-8 weeks of age. Support for this system is based on anecdotal evidence of increased rumen development (by the promotion of increased solid feed intake), and prevention of excess fat deposition. Conversely, aside from reducing animal welfare by maintaining calves' in a chronic state of hunger[2], the potential for optimal growth is greatly reduced.

Hypothesis

We hypothesised that Holstein bull calves given *ad libitum* access to MR would have higher growth rates but would not have significantly greater proportions of carcass fat than calves fed a restricted MR diet.

Methods

This study was nested within a larger study that recruited 100 Holstein heifer calves into one of two dietary groups at birth (Table 1). Nine Holstein bull calves were also assigned to each of the dietary groups. A further 3 bull calves were euthanased at birth, and 3 calves in both dietary groups were euthanased at 3, 9 or 12 weeks of age.

Table 1: Overview of husbandry protocols used for a) restricted MR and b) *ad libitum* MR feeding groups.

a) Restricted MR protocol	b) *Ad libitum* MR protocol
Bucket fed	**Teat fed from automatic feeder**
Individually housed for first 3 weeks, then grouped (max n=6 per group, age difference no more than 2 weeks)	**Group housed from birth (max n=6, age difference no more than 2 weeks)**
3 litres of MR fed 2 x daily	***ad libitum* MR allowance**
Weaning began at 8 weeks and MR intake was gradually reduced to 0 over a 1 week period	**Weaning began at 9 weeks of age and MR intake was progressively limited to 0 over a 3 week period**
***Ad libitum* access to water, hay and concentrate feeds from birth**	***Ad libitum* access to water, hay and concentrate feeds from birth**

Calves were systematically dissected immediately post-euthanasia. Body organs, gastro-intestinal tract (GIT), digesta and visceral fat were individually weighed (±0.1g). Carcasses were split longitudinally and the right half carcasses were stored at -20°C pending determination of tissue composition following spiral Computed Tomography (CT) (Somatom Esprit, Siemens, Germany).

Statistical analysis was carried out using STATA 12 (StataCorp, U.S.A.). Data was tested for normality and subsequently, Students t-tests were used to compare the composition of calves in both dietary groups at each age.

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Results

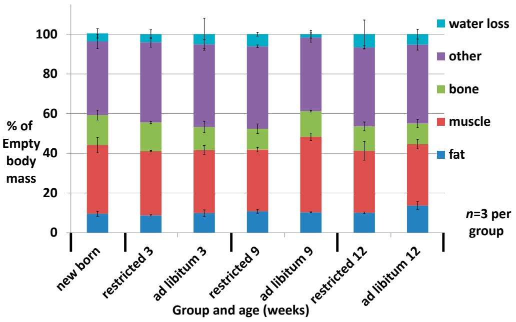

Figure 1: Carcass composition (mean ± s.d.) of newborn calves & all other calves at each age in both restricted & *ad libitum* fed groups. Empty body mass (bm) = Total bm (at time of death) – GIT contents.

Figure 1 shows no differences in carcass composition of all calves in the study. There was approximately 10.5% of fat in carcasses in all age groups.

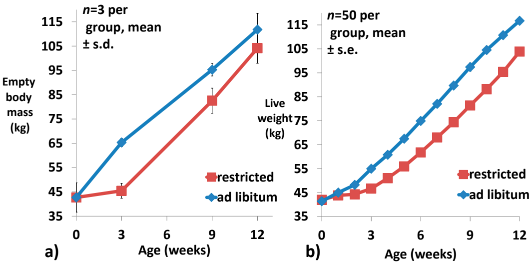

Figure 2: a) Empty body mass of bull calves at all ages in both groups (mean ± s.d.) and b) Total body weight of heifer calves in the larger study from 0 to 12 weeks of age (mean ±s.e.).

Calves in the *ad libitum* MR fed group had a higher empty body mass than calves in the restricted MR fed group at 3 (P=0.001), 9 (P=0.020) and 12 (P=0.150) weeks of age. The failure to demonstrate statistical significance in empty body mass between the two groups at 12 weeks of age is likely due to the small sample size. Data from the larger study indicated that *ad libitum* MR fed calves were significantly heavier from 3 weeks of age (P<0.001) than restricted MR fed calves.

Conclusions

This study would support *ad libitum* MR feeding of dairy calves. Benefits gained from this protocol include improved animal welfare and higher growth rates in early life. This can be achieved without compromising future health (heifer calves) or carcass quality (bull calves) resultant on increased adiposity. *Ad libitum* MR feeding should allow heifer calves to have an earlier age at first service and entry into the milking herd, and bull calves to achieve a higher carcass weight.

Fig 3: Image of one 'slice' from spiral CT of calf carcass

Workgroup 2

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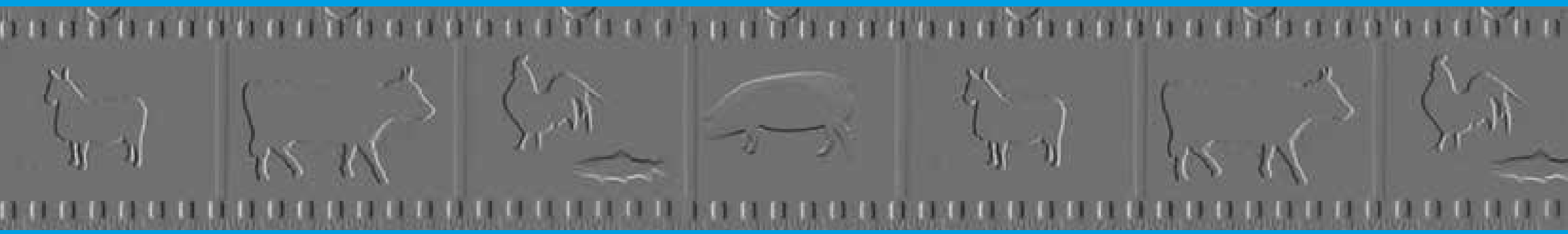

Preface to Workgroup 2

Milestones 2 and 6 of WG2 were the identification of relevant meat quality parameters for pig, beef, sheep and poultry. For this purpose a survey was performed and a questionnaire was sent to the different FAIM participants.

For this purpose a survey was performed and a questionnaire was sent to the different FAIM participants. The objective was to distribute the questionnaire to the different stakeholders of the production chain as well as to the research centres and universities of each of the participant countries. The aim of the survey was to identify the main meat quality attributes and the technologies used to determine them. A total of 106 questionnaires from 17 EU countries were collected: 34.9% for pig, 31.1% for beef, 17.9% for ovine and 9.4% for poultry (the rest were for rabbit, fish and game animals). Based on the results of the survey and following discussions with various meat scientists, a list of attributes by species was presented and discussed at the FAIM II conference.

The presentation was the focus of WG2 discussion, during which the various attributes were considered and consensus was reached that the list was agreeable to all FAIM participants present. The final list of relevant meat quality parameters is presented below. It is important to note that each quality trait is not necessarily a simple measure and the list provides general traits that can include a range of more detailed traits. For instance: water holding capacity includes drip loss and cooking loss among others, tenderness includes instrumental texture, trained panel and consumer assessment, and pH, colour and/or drip losses are relevant traits used for the classification of meat into the different quality categories in terms of pale, soft, exudative (PSE), dark, firm, dry (DFD), red, firm, non-exudative (RFN) and others.

PORK	**BEEF**	**LAMB**	**POULTRY**
pH	pH	pH	pH
Water holding capacity			Water holding capacity
Intramuscular fat	Intramuscular fat	Intramuscular fat	Intramuscular fat
Fat/muscle colour	Fat/muscle colour	Fat/muscle colour	Fat/muscle colour
Tenderness	Tenderness	Tenderness	
	Juiciness		Juiciness
Taste (taint)		Taste (taint)	
	Flavour		
Fatty acids composition	Fatty acids composition	Fatty acids composition	Fatty acids composition

Spectroscopic and imaging technologies to evaluate meat quality: a preliminary review

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Value for Industry

- Properties of raw and processed meat can be very variable. For the industry to implement technologies it is important to know the precision and potential for practical application, how the technologies can optimize their processes so as to improve economic benefits, to produce quality meat and meat products and to satisfy consumer demands.
- There are several technologies which are appropriate for the evaluation of meat properties, and some of them are non-destructive and suitable to be used on line. Other technologies need more development before its industrial implementation.
- Knowledge about the performance of each technology as well as the feasibility for its on-line use is of interest for the meat industry.

Background

Published works dealing with spectroscopic and imaging technologies have increased in the last decades due to the improvements not only in the technology *per se* but also in the systems to analyze high amount of data and images. A preliminary literature review of these technologies useful for the evaluation of raw and processed pig, beef, lamb and poultry meat quality was performed.

Why work is needed

A review of the currently available rapid, non-invasive technologies for the evaluation of the quality properties of meat and meat products is of interest to provide industry with information, which may be useful to improve their products and processes. The implementation of these technologies can also help to meet consumers' demands.

The technologies studied

The technologies that have been reviewed are nuclear magnetic resonance (NMR), ultrasound (US), computed tomography (CT), dual energy X-ray absorptiometry (DXA), video image analysis or computed vision system (VIA) and different spectroscopic technologies (near infrared - NIRS, RAMAN, hyperspectral imaging - HSI and microwave reflectometry - MW).

These technologies have been used to determine different meat quality attributes such as water-holding capacity (WHC), pH, intramuscular fat (IMF) and marbling, shear force (SF), sensory properties, colour and fatty acids, water content, salt, nitrates/nitrites, protein, ash, etc.

The calibration and validation of the different technologies have been performed using specific reference methods, which differed between studies. Also the performance is not always evaluated the same way, which makes the comparisons difficult. For this reason, the results are presented, for each technology and trait of interest (for different meat species), in terms of coefficient of determination (R^2).

The results obtained

In the last decade, around 17 works using NMR to estimate different meat quality traits have been published. All the studied applications, except those presented to determine IMF (R^2=0.8) in the technical note of Corrêa *et al*. (2009), would be difficult to be implemented on line at the industry due to the necessity of sample preparation.

US has also been used, either *in vivo* or post mortem, to estimate raw meat quality characteristics, mainly IMF, with the highest reported R^2 of 0.79 in beef (Aass *et al.,* 2009) and 0.92 in pork (Newcom *et al.,* 2002). Recently, Corona *et al.* (2013) also assessed textural changes in vacuum packaged sliced Iberian ham after high pressure treatment or cold storage.

CT has been used on fresh beef (Prieto *et al.,* 2010) to determine various quality parameters obtaining the best results in prediction of saturated fatty acids (SFA) (R^2=0.61-0.64) and monounsaturated fatty acids (MUFA) (R^2=0.66-0.76) fatty acids and IMF (R^2=0.71-0.76). CT also produced good results for the prediction of IMF (R^2=0.45-0.61) in sheep (Lambe *et al.,* 2008), pork loin (R^2=0.48-0.81) (Font-i-Furnols *et al.,* 2013) and dry-cured ham (Santos-Garcés *et al.,* 2014). Moreover, Fulladosa *et al.* (2010), Håseth *et al.* (2012), Santos-Garcés *et al.* (2010) and Vestergaard *et al.* (2004) used CT to estimate various meat quality parameters of dry cured ham such as salt (R^2=0.7-0.9), water (R^2=0.67-0.95), IMF (R^2=0.97) and water activity (R^2=0.83-0.95), showing a good ability of the technology to study salting and drying processes. Nevertheless, because of the characteristics of the medical CT devices, it is difficult to implement this technology on line at the industry. At present, efforts are being made to develop a device suiting industrial requirements.

Simpler X-ray based medical devices such as DXA could fulfill better industrial needs. DXA has been reported to be able to determine the chemical composition of pork and beef (R^2=0.5-0.9) (Brienne *et al.,* 2001). Recently, López-Campos *et al.* (2013) evaluated IMF of beef using this technology (R^2=0.43) and meat tenderness evaluation was also reported to be feasible (R^2=0.69) (Kröger *et al.,* 2006). Salt content (R^2=0.75) and fat content (R^2=0.51) of boned dry-cured ham has also been assessed using X-ray inspectors (Fulladosa *et al.,* 2013a).

VIA technology is mainly used on line to estimate carcass characteristics. Nevertheless, it can also be used (not always on line) to estimate IMF (R^2=0.22-0.74) or colour in chicken and turkey (Chmiel *et al.,* 2011), tenderness in beef (R^2=0.12-0.70; Li *et al.,* 1999; Sun *et al.,* 2012) or colour (R^2=0.52) and total viable count (R^2=0.64) (Lu *et al.,* 2000) in pork.

Since 2000 a number of publications have reported the use of NIRS to determine various meat characteristics and there are several relevant reviews (Prevolnik *et al.,* 2004; Prieto *et al.,* 2009; Weeranantanaphan *et al.,* 2011). NIRS can determine, in intact or minced meat, WHC, L*a*b*, SF, IMF, protein, moisture, fatty acids among others. Performance varies according to the analytical task; being related to sample preparation, and data treatment and analysis. This technology can be used on line although most of the works have been performed in laboratories, either with intact or minced meat. Recently, Prieto *et al.* (2013) have evaluated NIRS in the abattoir indicating reasonable estimation of polyunsaturated fatty acids (PUFA) R^2=0.78 for n-6, R^2=0.86 for n-3, R^2=0.67 for conjugated linoleic acid and R^2=0.67-0.78 for *cis* and *trans* MUFA. In dry-cured products, Collell *et al.* (2012) also studied salt and water content and reported the convenience of using this technology on line to control drying processes.

Interest in HSI technology has grown considerably from 2007 (Qiou *et al.,* (2007a,b) Naganathan *et al.,* (2008), Barbin *et al.,* (2012a,b,) (2013a); ElMasry *et al.,* 2012a; Wu *et al.,* 2012,). It has been used to estimate various pork, beef and lamb meat quality traits such as IMF (R^2=0.83 and R^2=0.94 for intact and minced meat respectively), WHC (R^2=0.77), colour L*a*b* (R^2=0.71-0.96), SF (R^2=0.77-0.83), sensory tenderness (R^2=0.53) and juiciness (R^2=0.49). Studies have been mainly laboratory based, but it would be possible to apply this technology on line. Indeed, a recent publication from Gou *et al.* (2013) showed the feasibility of an on line device to estimate the contents and distribution of water, fat and salt in packages of sliced dry-cured ham. HSI is an alternative to VIA or spectroscopy if more accurate results are needed and it can be used to fulfill meat industry requirements as reviewed by ElMasry *et al.* (2012b).

Since 2000, Raman spectroscopy has been used in pork, lamb and beef to estimate various meat characteristics. Results were promising, Wang *et al.* (2012) obtained a R^2 of 0.98 to predict chewiness, tenderness and juiciness and Olsen *et al.* (2007) R^2=0.83-0.98 to predict SFA and MUFA directly from fat tissue. This technology was not studied on line but recently, a portable device has been developed (Sowoidnich *et al.,* 2013). It has been found to be useful to evaluate the SF and cooking loss of sheep meat (Schmidt *et al.,* 2013) showing the potential of Raman spectra for the prediction of these quality traits in industrial environments.

MW spectrometry has been found useful for the determination of the presence of added water in different pork products (Kent *et al.,* 2002), to control pork meat salting process (Castro-Giráldez *et al.,* 2010), to identify pork meat quality (Castro-

Giráldez *et al.,* 2011) and beef meat traits (Clerjon and Damez, 2009). Time Domain Reflectometry (TDR) devices based on this technology can be built small to facilitate the on line measurement of meat properties for the industry (Sequid GmbH, Bremen, Germany). Fulladosa *et al.* (2013b) demonstrated the suitability of this device to evaluate salt (R^2=0.94) and water (R^2=0.95) percentages as well as a_w (R^2=0.98) in dry cured ham.

The scientific conclusions

- **Performance of different technologies in the determination of meat quality properties depends on the used technology and the trait. It is important to find, for each trait of interest the most appropriate technology in terms of accuracy, cost, efficiency and other requirements. The combination of technologies could allow a better determination of meat properties.**
- **Some technologies are suitable to be used on line although further work is needed to prepare them for this purpose in order to 1) avoid or minimize pretreatment of the samples, 2) allow a continuous measurement in the carcass or the meat, and 3) get an appropriate production speed as well as other necessities of the industry.**
- **The value of the technologies is not only in industrial use, these methods can serve for screening purposes or for breeding programs where lower accuracy of rapid method as compared to analytical values is compensated by high number of results.**
- **The chemometrics used to work with images and data from these technical devices are complex and it influences considerably the performance obtained. Thus, it is also important to work in the automation of data/image analysis to allow a good implementation of the technology on line and to provide quick results.**

The next steps

This paper is just a short overview of existing non-destructive technologies able to determine meat quality traits. In the follow up, the review will be completed and presented to the scientific and industrial community with information adapted to their requirements. Firstly, the most important attributes for each meat species have to be determined and then, from the review, the suitability of the technologies to determine each of them. Advantages and drawbacks of each technology will be also discussed.

Acknowledgements

COST Action FAIM FA1102 "Optimising and standardising non-destructive imaging and spectroscopic methods to improve the determination of body composition and meat quality in farm animals" is greatly acknowledged.

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Effect of dietary fat level on the gross fatty acid profile of pork back fat: Raman spectroscopic study

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Value for Industry

- Raman spectroscopy has value for the meat industry:
- It is easy, fast, non-destructive and not sensitive to water. Thus, the meat industries can make measurements and classify carcass and/or product rapidly.
- Raman spectroscopy is becoming a promising technique to be used in the meat industry as it can predict fatty acid composition in pork fat with a very good correlation.

Background

Fatty acid composition of meat is becoming a concern for consumers due to the health related issues. Different strategies such as cross breeding and gender (Alonso *et al.*, 2009) and feeding trials (Pascual *et al.*, 2006) have been developed with the intention to improve the fatty acid profile in the pork meat and meat products. Dietary fat level in a feed is one of the factors which determine the fatty acid profile of pork back fat. It is therefore important for the slaughter house to determine the fatty acid composition of the fat as it also affects the quality of meat products during processing. Such interest is leading the call for the development of an easy and rapid technique which could be used in the industry. Raman spectroscopy is a potential technique to be used in the meat industry. It gives information at the molecular level.

The principle is that a sample is radiated with a monochromatic visible or near infrared light from a laser and a vibrational transition takes place in the ground electronic state of molecules. As a result, the vibrational energy level in the molecule reaches to a high energy collision state for a short time and go down to a lower energy state by emitting a photon which has a lower frequency compared with the laser light (Herrero, 2008). The advantage of Raman spectroscopy is that it is fast, insensitive to water and special sample preparation is not required. This technique can be used in the meat industry to make classification of fat when it is going to be used as a raw material for processing. Hand-held instruments are already in use for research on meat quality (Schmidt *et al.*, 2013) and useful preliminary results in relation to pork back fat have also been reported (Lyndgaard *et al.*, 2012).

Why work is needed

The fatty acid profile of pork fat is becoming an issue for the consumers and the fatty acid composition also affects the quality of processed products. The objective of this study was to use Raman spectroscopy to characterize fatty acid composition of pork back fat with the aim of predicting the iodine value (IV) (which is a measure of the amount of unsaturation in fatty acids) of the back fat from pigs fed with different dietary fat level. This could help the slaughter house to make a fast measurement in the slaughter line and classify a carcass based on its quality for further processing.

The methods used

Pigs were fed with different levels of dietary fat. The finished pigs were slaughtered in a commercial slaughter house (Danish Crown, Denmark). From the left side of the carcass, the *Longissimus* muscle between the 2nd and 4th lumbar vertebrae was removed from the carcass and the back fat was sampled at 24 h post mortem. For Gas Chromatography (GC) measurement, samples were vacuum packed and stored at -20 °C until use.

For Raman microscopy measurement, the samples were vacuum packed and stored at 0 °C for 3 days then small area (2*3 cm) was sampled and the inner and outer layers were separated. Raman measurement was made on the new cut surface of slices from each layer. Raman and Gas Chromatography measurements were made according to the literature (Lyndgaard *et al.*, 2012) with some modifications. In brief, Gas chromatography was used to analyze the fatty acid profile of the back fat (homogenous sample of inner and outer fat layers) samples.

Iodine Value (IV) was calculated from the fatty acid profiles in order to determine the degree of unsaturation of the adipose tissue. Raman spectra were collected using a Raman instrument (Kaiser Optical Systems Inc, Michigan, 131 USA) equipped with a 785 nm near-infrared diode laser (Invictus, Kaiser Optical Systems Inc., 132 Michigan, USA). Raman spectra (1800-200 cm^{-1}) were collected separately from the inner and outer layers using the average spectrum of 16 scans each with an exposure time of 1 s. Average of the Raman spectra both from inner and outer fat layers of each pig was used for analysis in order to be able to predict the calculated IV from the GC measurement. The Raman spectra were preprocessed using Standard Normal Variate (SNV) before performing Principal Component Analysis (PCA) and Partial Least Square Regression (PLSR) analysis.

The results obtained

Iodine Value (IV) of the back fat was increased (minimum 61 and maximum 81) when the dietary fat level was increased. This result showed that the level of the dietary fat intake had an effect on the degree of unsaturation of the back fat. Principal component analysis (PCA) was performed on the preprocessed Raman spectra and there was a clear discrimination among the spectra when the score plot was coloured according to the measured IV (Figure 2 left). Principal component-1 was able to explain about 86.63 % of the variation among the spectra. From the loadings plot, it was found that the main contributors for the discrimination among the spectra were the bands originating from C–C in-phase stretch (1128 cm^{-1}) and >CH twisting (1296 cm^{-1}) (Figure 2 right) showing that the samples with smaller IV had a higher amount of saturated fatty acids. As mentioned above, average spectra from both the inner and outer layers of the back fat of each animal were calculated and were used to predict the IV obtained from the reference method (GC). Partial Least Square Regression (PLSR) was performed and good correlation between the average Raman spectra and IV of each animal was found (R^2=0.71) with a root mean square error of cross validation (RMSECV) of 2.7 using only 2 PLS components (Figure 3 left). The regression vector for the PLS model (Figure 3 right) showed that there was a higher contribution for the model from the C–C in-phase stretch (1128 cm^{-1}) and >CH_2 twist (1296 cm^{-1}).

Figure 1. Raman microscope.

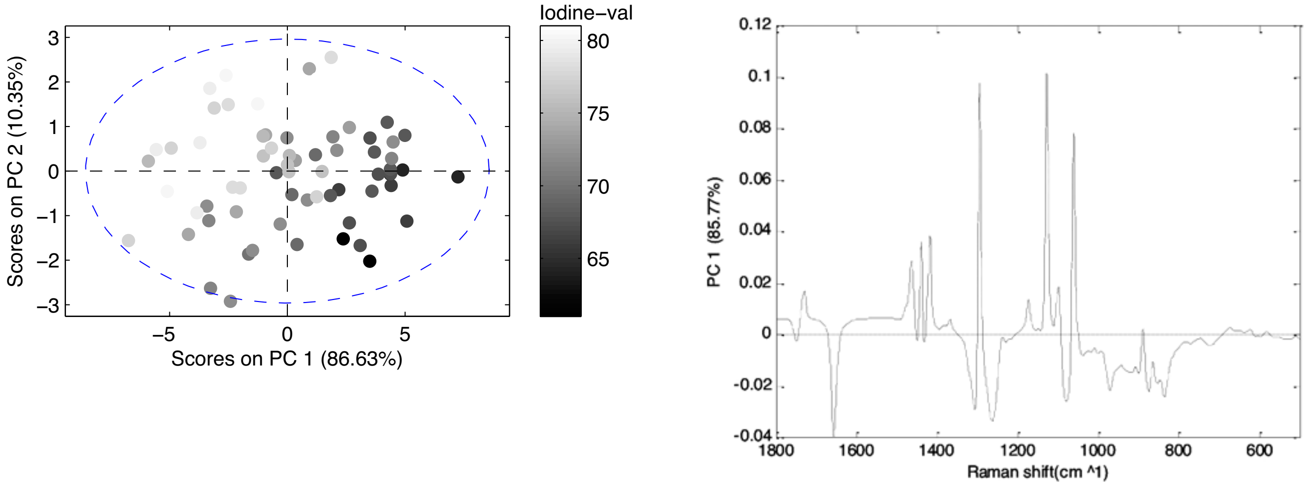

Figure 2. Principal component analysis (PCA) of Raman spectra (1800-500 cm^{-1}) from pork back fat. PC-score plot (PC-1 vs -2) coloured according to iodine value (left). PC-1 and -2 explained 96.98 % of the variation in the Raman spectra (n=65), and PC-loadings plot for PC-1 (right).

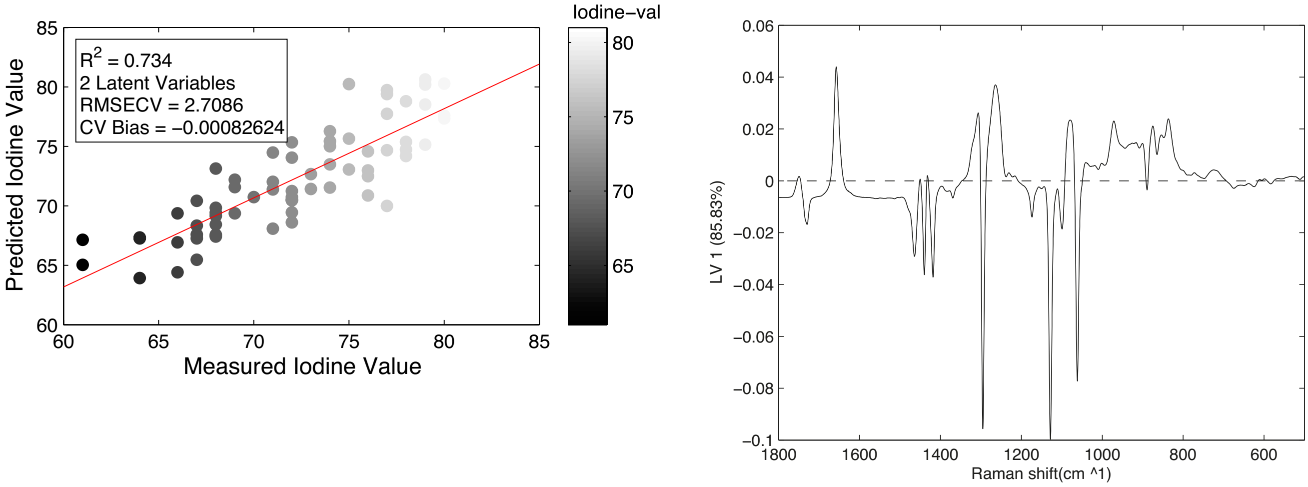

Figure 3. Measured vs. predicted iodine value (left) and Latent variable-1 (right). Raman spectra (1800-500 cm^{-1}). Coloured according to iodine value. X-data-preprocessed using standard normal variate, Y-data-autoscaled (n=65).

The scientific conclusions

Raman spectroscopy is a promising technique to be used as a fast technique for classification of carcasses based on the gross fatty acid profile. It was possible to predict the IV with a good correlation though the Raman measurement was made on the separate layers and the spectra from both layers were averaged, and the GC measurement was made on a homogenous sample from inner and outer layers of each pig. This good correlation shows the potential of Raman spectroscopy to predict the IV.

The next steps

The current study was performed using a Raman microscopy (with a 10x objective). The next step will be to use a probe and/or hand-held Raman in order to study the consistency of such good results. This will help to make an evaluation on the applicability of the technique at the slaughter house either at-line or on-line. In this experiment, the GC measurement was made on homogenous samples of the inner and outer layers but it would be useful to make the GC measurement also on the separate layers but the same samples and compare the strength of the model with the current results.

Acknowledgements

This study was funded by the University of Copenhagen and Danish Meat Research Institute (DMRI).

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Prediction of intramuscular fat in Texel lamb loins using different x-ray computed tomography (CT) scanning techniques

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Value for Industry

- Development of a fast, non-destructive analysis tool to accurately measure meat quality (MQ) and meat eating quality (MEQ) traits *in vivo.*
- *In vivo* measurement methods could allow establishment of informative estimated breeding values (EBV's) for intramuscular fat (and associated MEQ traits).
- EBVs for new CT traits could facilitate continued improvements in growth and carcass composition, whilst maintaining or improving lamb MQ and MEQ.
- Enabling selection for lean carcass growth without compromising MEQ in lamb breeding programmes.

Background

Computed tomography (CT) is a sophisticated, non-invasive, diagnostic tool initially developed for use in human medicine and over the last few decades has been adopted for use in animal breeding, offering comprehensive and reliable information on body composition and, more recently, aspects of meat quality (Bünger *et al.,* 2011). During single-slice scanning, x-rays are used to generate cross-sectional, two-dimensional images of the selected region of the body. Each image is produced by rotation of the x-ray tube 360° around the subject. Attenuation of radiation through the tissues can then be measured, which changes with differing tissue density. From this, images are then produced using the attenuation values represented by a variance of greyscale. These images can be analysed by specialist software to quantify areas and densities of different tissues in the total body, or in specific regions.

Spiral CT scanning (SCTS) is a relatively new imaging technology capable of producing a series of images in a single contiguous scan, at intervals of as little as 1 mm apart. During SCTS, the x-ray source moves freely in a continuous rotation while the scanner table moves smoothly in one direction. The advantage of this procedure is that multiple images may be taken at increased speeds, resulting in higher resolution acquisitions in less time, enabling 3 dimensional imaging and measurement of volumes, weights and density of tissues.

Why work is needed

CT scanning is a routine measurement method used in several selective breeding programmes for sheep in the UK to accurately estimate carcass composition. It provides the potential to quantify both total carcass tissue weights (muscle, bone, fat) and intramuscular fat (and associated meat quality traits) *in vivo*. Due to increased consumer awareness in aspects of meat quality it is becoming increasingly important to retailers and therefore animal breeders. It is necessary to understand if additional meat and carcass quality parameters can be assessed using CT or SCTS, and whether these predictors can also be incorporated into processing procedures and/or breeding programmes, allowing continued improvements in growth and carcass composition, while maintaining or possibly improving aspects of meat quality and meat eating quality.

The aim of this experiment was to investigate the ability of SCTS to predict intramuscular fat (IMF) in the loin muscle of Texel lambs *in vivo*.

The methods used

Spiral CT images at intervals of 8 mm were selected from the loin region of Texel lambs pre-slaughter (n=439). Mean age at slaughter was 149 days (SD 21.53, range 99-234 days), mean live weight at slaughter was 34.33 kg (SD 5.5, range 19.71-52.2 kg). The majority of lambs were slaughtered 4-8 days after CT scanning (n = 216), with a number slaughtered 13-15 days after CT scanning (n = 72). The remaining lambs were slaughtered 32-33 days after CT scanning (n = 159), to allow for a 30 day withdrawal period from the CT sedative and subsequent taste panel analysis, which formed part of the wider study. Carcasses were subjected to high voltage electrical stimulation, chilled and aged for between 7-9 days and dissected, removing muscles from the right side of the carcass (*M. longissimus lumborum*), which were vacuum-packed and frozen. Samples were later thawed and chemical IMF percentage was measured using petroleum ether (B.P. 40-60°C) as the solvent in a modified Soxhlet extraction.

For CT image analysis, selection of the first SCTS image in the sequence of spiral images was based on the appearance of the 7th lumbar vertebra and the final image was selected as the first image in which the 1st lumbar vertebra was no longer visible (Figure 1). Three cross-sectional single-slice reference scans were also taken in the chest region at the 8th thoracic vertebra (TV8), the loin region at the 5th lumbar vertebra (LV5) and in the leg region at the ischium bone (ISC), which are the three "reference" scan images commonly used in commercial sheep CT scanning to calculate predictions of carcass tissue weights.

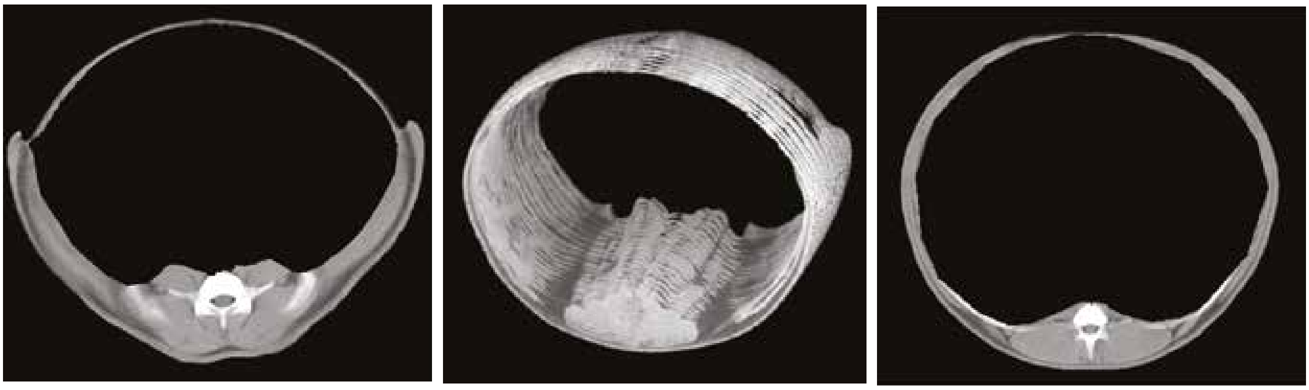

Figure 1. First image where LV7 appears (left), last image where LV1 is no longer visible (centre) and 3D rendered stack of selected images (right).

Image analyses were performed to separate carcass from non-carcass tissues (Glasbey and Young, 2002) and the density of each pixel in the carcass portion was allocated to fat, muscle or bone, according to density thresholds using Sheep Tomogram Analysis Routines (STAR) software (Mann *et al*., 2003). Areas (mm^2) and average densities (Hounsfield units - HU) of each tissue (muscle and fat) in each 2D image were calculated, as well as standard deviations for the density values of all pixels allocated to each tissue. A novel soft tissue density and its standard deviation was also calculated, combining the information from both fat and muscle tissue densities. The 3D loin images were used to calculate weighted CT density (average tissue densities across all images in the spiral scan, weighted for tissue area in each image), and volume (mm^3). The resulting spiral CT parameters included weighted muscle and fat densities (MD, FD), their standard deviations (MSD, FSD), weighted soft tissue densities (STD) and standard deviation (STSD) and calculated muscle and fat tissue volumes (MA, FA). The CT parameters measured from the 3 reference scans were defined in the same way. Total carcass fat (Pr_Cfat), as a measure of subcutaneous and intermuscular fat in the entire carcass, was also predicted using a breed-specific prediction equation developed from previous research (Macfarlane *et al.,* 2006). Animals included in the data set without full CT information (n=2) and animals with no IMF data were removed (n=5), and finally animals identified as outliers (IMF% >3SD from the mean) were also removed (n=3). Number of days from CT to slaughter was tested for significance, but showed no significant effect on IMF levels, so was therefore not included in the statistical analysis. Sixteen models were tested in the analysis to predict IMF, using different combinations of (i) spiral loin CT information, and (ii) spiral loin CT information plus single-slice CT information from the TV8 and ISC images. Different models added CT measurement traits in a progressive manner. Statistical analyses used simple, multiple and generalized stepwise linear regression in Genstat14™ (Payne *et al.,* 2012).

Models were then tested for significant differences using their correlation coefficients and applying Fisher's Z transformation (Rasch *et al.,* 1978) (√Adj R^2) where Adj R^2 = 1- (*Residual MS/ Total MS*), the *Total MS* is the sample variance, and the *residual MS* is an estimate of, the variance of a value of Y given the set of X's. The resulting statistic is less biased than R^2, and a better measure to use when comparing models with different numbers of predictors. Firstly models were tested against the single best predictor (Pr_Cfat) to identify models significantly more accurate in their prediction ability, and subsequently these results were used to identify models which did not differ significantly from the single best model.

The results obtained

CT predicted carcass fat accounted for a moderate amount of the variation in IMF (Adj R^2 0.5). There were twenty one models from the thirty models tested (fifteen in each of the image analysis methods, (i); spiral loin CT information and (ii); spiral loin plus reference CT information) with statistically significant improvement in accuracy of prediction when compared to CT predicted carcass fat as a single predictor ($p<0.05$). Prediction accuracy was significantly increased to Adj R^2 0.62 with the inclusion of average muscle density weighted and averaged across all images selected from the spiral scans (22 – 29 images). The single best model identified was using information from both the loin spiral and the TV8 and ISC scans (Adj R^2 0.71) (Table 1) and from this all models from both image analysis methods that had statistically similar prediction ability were identified using the method previously described. This resulted in thirteen models being identified as similar in their prediction accuracies with Adj R^2 ranging from 0.65 – 0.71 (Table 1).

Table 1. Linear regression models between IMF and CT parameters, with adjusted coefficient of determination (R^2) and residual mean square error (RMSE)

Maximum Model	**Spiral only**[1]		**Spiral + Reference**[2]	
	Adj R^2	RMSE	Adj R^2	RMSE
A – Pr_Cfat	0.50	0.47	0.50	0.47
B – Pr_Cfat+MD	0.62	0.41	0.63	0.41
C – Pr_Cfat+FD	0.52	0.47	0.52	0.46
D – Pr_Cfat+MA	0.56	0.45	0.59	0.43
E – Pr_Cfat+FA	0.57	0.44	0.59	0.43
F – Pr_Cfat+MD+FD	0.65*	0.40	0.68*	0.38
G – Pr_Cfat+MA+FA	0.59	0.43	0.60	0.42
H – Pr_Cfat+MD+MSD	0.65*	0.40	0.67*	0.38
I – Pr_Cfat+FD+FSD	0.53	0.46	0.54	0.46
J – Pr_Cfat+MD+MSD+FD+FSD	0.68*	0.38	0.70*	0.37
K – Pr_Cfat+MD+MSD+FD+FSD+FA	0.68*	0.38	0.70*	0.37
L – Pr_Cfat+MD+MSD+FD+FSD+MA+FA	0.68*	0.38	0.71*	0.36
M – Pr_Cfat+STD	0.55	0.45	0.64	0.40
N – Pr_Cfat+STD+STSD	0.56	0.45	0.65*	0.40
O – Pr_Cfat+STD+STSD+FA	0.58	0.44	0.66*	0.39
P – Pr_Cfat+STD+STSD+FA+MA	0.60	0.43	0.66*	0.39

[1] Using information from selected and weighted spiral CT results

[2] Using combined information from selected and weighted spiral CT parameters and reference scan CT parameters (TV8 and ISC)

*√Adj R^2 are not significantly different when tested using Fisher's Z transformation

The scientific conclusions

Spiral CT scanning of live Texel lambs can therefore provide an accurate prediction of IMF in the loin, using information from selected spiral images across the loin region (LV1 – LV7) with a maximum Adj R^2 0.68. The accuracy of prediction can be further increased (although not significantly) by including information from two additional single-slice anatomical scans, positioned at two reference scan sites, achieving Adj R^2 0.71.

However, this method of image capture and analysis did not achieve significantly higher accuracies than similar models employing CT information from only the single-slice images from three reference scan sites, including CT predicted carcass fat, fat and muscle densities and their standard deviations, which achieved an Adj R^2 0.67 (Clelland *et al*. Unpublished).

The next steps

The most accurate prediction models identified from the current analysis will be calibrated using a subset of the entire data set, and cross validated using the remaining independent portion of the data, in order to obtain the most robust and transferable model or models. The resulting method, be it from reference imaging, spiral imaging or a combination of both, will be applied to a powerful data set to estimate genetic parameters, the aim being to allow the maintenance or improvement of IMF genetically, without compromising important production traits, including carcass quality.

Acknowledgements

The authors would like to acknowledge the support of EBLEX, QMS and HCC. Acknowledgement of technical support in the chemical analysis of IMF goes to staff at the University of Bristol.

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Use of computer tomography (CT) to predict chemical intramuscular fat (IMF) in dissected lamb loins

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Value for Industry

- The prediction of IMF during processing of retail cuts in a non-destructive manner, independent of any fixed effects.
- The potential to provide a valuable commercial tool to maximise levels of eating quality and the foresight to target certain markets.
- Provide information on meat quality to feed into breeding programmes.
- Non-destructive prediction of eating quality which could provide information as a basis to reward commercial farmers for meat quality.

Background

In different livestock species, meat eating quality (MEQ) traits such as flavour, tenderness and juiciness are known to be linked to fat levels. This association of MEQ attributes and fat levels in meat are largely due to the positive association with intramuscular fat (IMF) (Savell and Cross, 1988). X-ray computed tomography (CT) can measure total fat, muscle and bone *in vivo* in sheep and CT predictions of carcass composition have been used in commercial UK sheep breeding programmes over the last two decades (Bünger *et al.,* 2011). Together with ultrasound measures of fat and muscle depth in the loin region, CT measured carcass fat and muscle weights have contributed much to the success of breeding for leaner carcasses and led to higher selection responses (Moore *et al.,* 2011).

Previous research has not only demonstrated that *in vivo* CT can predict or measure total carcass fat, but it can also provide measurements of the average muscle density, which has been shown to be a good predictor of IMF (Karamichou *et al.,* 2006; Young *et al.,* 2001). The application of CT technology in primal or retail cuts of lamb has not been investigated as thoroughly. The use of spiral CT scanning (SCTS) and the development of multi-object scanning may provide additional advances in the application of such technology at the time of processing. Multi-object spiral CT scanning provides high resolution CT images in a cost effective way, generating 3D images and precise measurements of tissue volumes and weights of several objects at the same time. Although there is continued investigation into the prediction of meat quality traits in lamb using *in vivo* CT, using both information from reference scanning and 3D spiral scanning, relatively few studies have focussed on primal or retail cuts.

Why work is needed

The provision of meat quality assurances from meat processors is certainly of interest to both the consumer and producer. For the consumer, an assurance of certain minimum levels of meat quality traits should increase confidence at point of purchase and omit requirement for visual evaluation by consumers. For the producer, knowledge of meat quality aspects may be fed into breeding programmes and future payment systems would have relevant information to draw from to reward producers on meat quality. All these aspects would enable the processor to target certain markets and better evaluate markets and consumer preferences. The ability to measure lamb meat quality in a non-destructive and safe manner, maintaining the integrity of the product, is important for any such system to be applied in industry.

The aim of this experiment was to investigate the ability of CT to predict intramuscular fat (IMF) in a retail processed cut of meat (*M. longissimus*) using only CT derived information.

The methods used

Two hundred and two rear right side lamb loins (Lumbar vertebra 1-7) were removed post-slaughter, vacuum packed, chilled and aged for seven days on the bone. After this time the bone was removed, and the loin's were re-packaged and frozen for transportation to the SRUC/BioSS CT unit, Edinburgh, UK for CT scanning.

The front portion of these same 202 loins (thoracic vertebra region) were split into three separate pieces 3 days post-slaughter, immediately vacuum packed and frozen for further analytical testing, as part of the wider study, including IMF estimation using the direct bimethylation method described by Lee *et al*. (2012). The section of the loin that included the last rib (14th thoracic vertebra) was thawed for 14 hours, visible fat and connective tissue were removed and 1cm^3 samples were cut. Chemical IMF (chem_IMF) was calculated as the sum of the major fatty acids in a fatty acid profile (mg/100g fresh weight).

The rear right loins (lumbar vertebra region), which were transported frozen to the SRUC/BioSS CT unit in Edinburgh, were thawed over a period of approximately 12 hours under refrigeration, over a 10–day period, in six batches of 30 samples and one batch of 22 samples. Loins were uniformly orientated and positioned on a multiplex scanning frame and spiral CT scanned (contiguous scans at 8mm intervals) in batches of six (Figure 1). Each batch was scanned at two separate x-ray intensities (80kV and 130kV). The purpose of this was to investigate whether images produced at 80kV would increase the contrast of voxels within the soft tissue, compared to the standard 130kV intensity, and therefore improve on the accuracies of prediction in IMF following a similar approach by Stubjkjaer-Schubert *et al.* (2009).

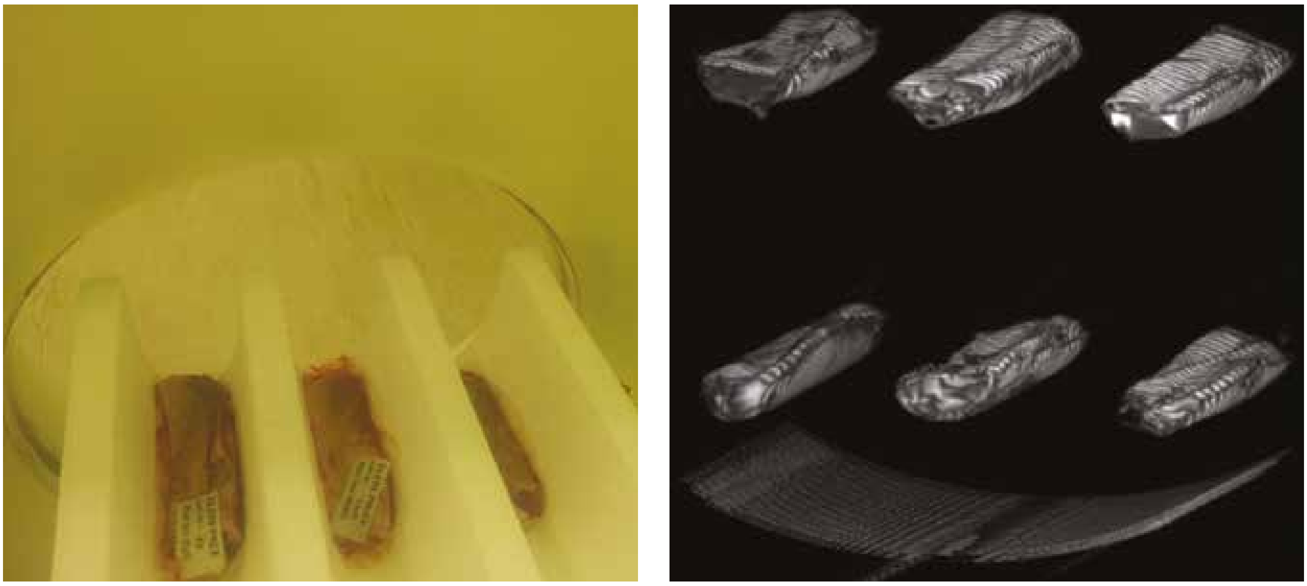

Figure 1. Dissected loins orientated and positioned in the multiplex frame (left), 3D rendered image of multiplex scanning (right).

CT images were segmented using a multi-object animal tomograph analysis routine (ATAR) developed at BioSS/SRUC (Figure 2). Each pixel in each image was allocated as fat or muscle, using previously developed density thresholds, specific to the analysis of images obtained from carcasses, primal cuts and dissected muscles (fat = -244 to 24 HU, muscle = 26 to 204 HU). The CT density results were then weighted by area in each image and averaged over the spiral series images (26-30 images per loin, average = 28 images).

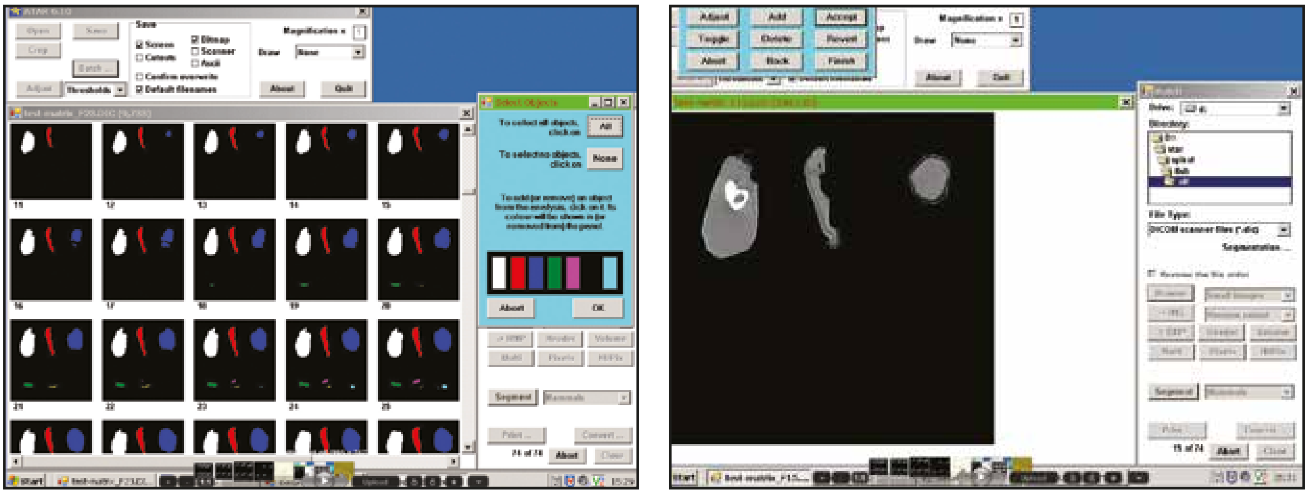

Figure 2. Selection of the scanned objects using ATAR software (left), adjusting the boundary of a selected object (right).

To predict the chem_IMF from CT measured traits, the statistical analysis included the weighted CT density information from each spiral loin scan, this included: loin weight at CT scanning (Ct_wt) calculated from the CT derived volume and weight (g/cm^3) of the soft tissue (muscle and fat); weighted muscle and fat densities (w_md, w_fd) and their standard deviations (w_msd and w_fsd); weighted soft tissue densities (combining the density ranges between fat and muscle) and standard deviation (w_std, w_stsd); and the proportion of voxels allocated as fat (Pr_F_vox). Nine models were tested in the analysis, using different combinations of CT variables in each maximum model. Models containing three or more variables were analysed using generalized stepwise linear regression in Genstat14™ (Payne *et al.,* 2012) optimising the use of predictor variables within the more complicated models, while simpler models containing a maximum of two predictor variables were analysed using multiple linear regression. Models were then tested for significant differences using their correlation coefficient ($\sqrt{\text{Adj } R^2}$) and applying standard methods.

The results obtained

Chem_IMF% ranged from 1.27% to 4.71% with a mean of 2.49% and a coefficient of variation of 22.94. Table 1 shows the regression parameters (Adj R^2 and RMSE) of the predictive models obtained from multiple and generalised stepwise linear regression analysis from the maximum models tested. Using CT calculated loin weight and muscle density at an intensity of 80kv resulted in very poor prediction of IMF% (Adj R^2 0.04; model A), with similar poor results using fat density and combining muscle and fat density (Adj R^2 0.06; models B and C). The use of soft tissue density and its standard deviation resulted in similar prediction accuracies (Adj R^2 0.04, 0.05; models G and H). However, the use of CT calculated loin weight, average muscle density across the loin and the standard deviation of muscle density resulted in a significant increase in accuracy (Adj R^2 0.18; model D). A further slight increase in accuracy (Adj R^2 0.20; model I) was achieved when the proportion of voxels allocated as fat was included in the model. No significant improvement was found using results obtained from the higher CT intensity (130kv). The maximum accuracy of prediction achieved was an Adj R^2 of 0.14 at 130kv.

Table 1. Linear regression models between chem._IMF and CT tissue parameters, with adjusted coefficient of determination (Adj R^2) and residual mean square error (RMSE)

		80kV		130kV	
	Maximum Model	**Adj R^2**	**RMSE**	**Adj R^2**	**RMSE**
A	Ct_wt+w_md	0.04	0.56	0.04	0.56
B	Ct_wt+w_fd	0.06	0.56	0.07	0.55
C	Ct_wt+w_md+w_fd	0.06	0.56	0.07	0.55
D	Ct_wt+w_md+w_msd	0.18*	0.52	0.13*	0.53
E	Ct_wt+w_fd+w_fsd	0.06	0.55	0.07	0.55
F	Ct_wt+w_md+w_msd+w_fd+w_fsd	0.18*	0.52	0.14*	0.53
G	Ct_wt+w_std	0.04	0.56	0.04	0.56
H	Ct_wt+w_std+w_stsd	0.05	0.56	0.05	0.56

*√Adj R^2 are not significantly different when tested using Fisher's Z transformation

The scientific conclusions

In this study, CT was unable to predict IMF content of dissected lamb loins with acceptable accuracy. However more promising prediction accuracies (R^2 0.33 - 0.44) have been reported in previous studies using a small sample of 30 dissected loins (Lambe *et al.*, 2010). In the same study, prediction accuracies varied when using different scanners (R^2 0.05 – 0.12). Live animal assessments use entire carcass images (including other fat depots such as subcutaneous and intermuscular fat), and therefore the poorer results obtained during the current study may suggest that there is additional information within these other carcass portions and fat depots which provide increased accuracies in prediction of IMF. Another factor may be that the composition of the muscle *in vivo* and post processing changes, in terms of the density of soft tissue, which results in the reduction of prediction ability when using density measures of processed retail cuts of meat. This may be further supported by substantially improved prediction accuracies during a separate study where virtually dissected loins from live animal scans resulted in accuracies of 62% (Clelland *et al*. Unpublished)

The next steps

Further research will be carried out investigating the prediction of IMF in primal cuts of lamb, including additional fat depots (subcutaneous and intermuscular) to determine the role that these fat depots may play in the accurate prediction of IMF in retail or primal cuts. Comparative data sets are available to investigate the difference in accuracy when using virtually-dissected images from live animal scanning to isolate tissue included in both retail and primal cuts, alongside CT scan data from butchered primal cuts and fully dissected loins.

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Use of Near Infrared Spectroscopy (NIRS) as prediction tool for the quality of fresh pork for cooked ham production

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Value for industry

- Near Infrared Spectroscopy (NIRS) is a fast and objective measurement which can be implemented in the production line of cooked hams in order to obtain batches with a more uniform quality.
- Since the purchase of fresh ham is about 70% of the total production cost, NIRS prediction of the technological quality of the fresh ham can ensure a better utilization of the raw material when processing high quality cooked ham, resulting in lower economic losses.
- Destructured zones are caused by a higher protein denaturation in the fresh meat. As most of the muscles sensitive to protein denaturation are present in the core of the ham, NIRS fiber optics can provide measurements at those muscles located in the depth of the ham.
- NIRS as prediction tool for the quality of fresh pork can allow a more efficient processing of cooked hams with lower slicing losses and a decreased volume of non-consumable products.
- An accurate classification of raw materials gives the meat processing industry the opportunity to decide at the beginning of the production line whether fresh hams can be used for the production of high quality cooked hams or certain technological additives need to be added.

Background

During the production of cooked ham, texture defects are often noticed (Vautier *et al.*, 2004; Hugenschmidt *et al.*, 2007). Destructured zones in cooked ham are estimated to be present in up to 20% of the cooked hams (Franck *et al.*, 1999) and are described by the meat industry as ‘soft zones, having an abnormal pale color, with a high loss of moisture’ (Hugenschmidt *et al.*, 2009). These destructured zones are more frequently observed when high quality cooked hams are produced, i.e. with a minimum of added technological aids during the process. The destructured zones are characterized by a higher myofibrillar fragmentation compared to normal meat (Hugenschmidt *et al.*, 2009), occurring due to a rapid glycolysis and slow temperature decrease post mortem. As most destructured zones are caused by a higher protein denaturation in the fresh ham and the muscles sensitive to protein denaturation are situated in the core of the ham, i.e. *Semimembranosus* (SM), *Adductor* (AD) and *Biceps femoris* (BF), defects are not visible during visual control of the fresh ham. Therefore, a fast and objective measurement, at the sensitive places in the core of the ham, is needed to give information on the technological quality of the fresh ham, and thus to predict its suitability for producing high quality cooked ham.

The aim of this study was to evaluate the use of NIRS as classification tool in order to classify fresh hams into different classes of quality, predicting the quality of the fresh hams for cooked ham production more accurately.

Importance of this study

As consumers increasingly pay attention to the quality of food products, the presence of structural defects in cooked hams is still a major concern for the processing meat industry. Destructured zones in the cooked ham are often correlated with lower yields, slicing problems, higher moisture loss and higher syneresis when MAP-packaging is performed. Concerning biochemical properties, destructured zones show strong similarity to those of pale, soft and exudative (PSE) meat (Laville *et al.*, 2005). Using extreme PSE fresh ham as well as ham with intermediate PSE characteristics can lead to cooked ham with a lower and non-acceptable end quality. Although efforts are done to separate pork with PSE characteristics and to process PSE meat to obtain final products with acceptable properties (Lesiów & Xiong, 2013), currently used objective and subjective methods are still not selective enough to distinguish intermediate PSE categories from good quality meat. As it is generally accepted that final pH values of PSE meat, measured 24 hours after slaughter, are quite similar to those of normal quality meat, pH measurements done at the reception of the meat factories are not satisfactory to detect meat with (intermediate) PSE characteristics. Hence, a more reliable technique is needed to detect (intermediate) PSE fresh ham more accurately, so that economic losses are minimized for the pork processing industry.

Materials and methods

A total of 55 fresh hams were selected at 3 different slaughterhouses based on pH and temperature values measured 30 minutes post mortem in the SM muscle. Since Balac *et al*. (1998) reported that the prevalence of destructured zones is positively correlated to a low pH_{30min}, some fresh hams with a pH_{30min} lower than 5.8 were selected to increase the probability of hams with inferior meat quality. After selection, fresh hams were transported to the laboratory and further stored at 4°C. At 24 hours post mortem pH and electrical conductivity, analyzed by a Pork Quality Meter (PQM), were measured on the raw, intact hams, in particular at the transition zone between SM and AD muscle as well as on the BF muscle. After deboning the hams, pH, PQM and HunterLab color values were measured on the different muscles. Sarcoplasmic and myofibrillar protein solubility were determined at level of the transition zone between SM and AD muscle according to Claeys *et al*. (2002); drip loss was analyzed according to Honikel (1987). Infrared spectra were collected using NIR technology equipped with a fiber optics probe over the range 4000-12000 cm^{-1}. Per sample, 128 scans with a resolution of 16 cm^{-1} were averaged. On the intact hams, 2 types of NIR measurements were carried out 1) on the surface of the BF muscle, 2) at the transition zone between SM and AD muscle where an incision was made to collect the NIR spectrum at 8 cm of depth. On the deboned hams, NIR spectra on both BF muscle and transition between SM and AD were recorded at the surface. Afterwards, raw hams were cooked in a pilot plant after which they were sliced and visually classified according to whether destructured zones were present (= normal quality) or not (= inferior quality). Figure 1 shows an example of both normal and inferior quality hams, visually classified according to the appearance of destructured zones in the sliced cooked ham.

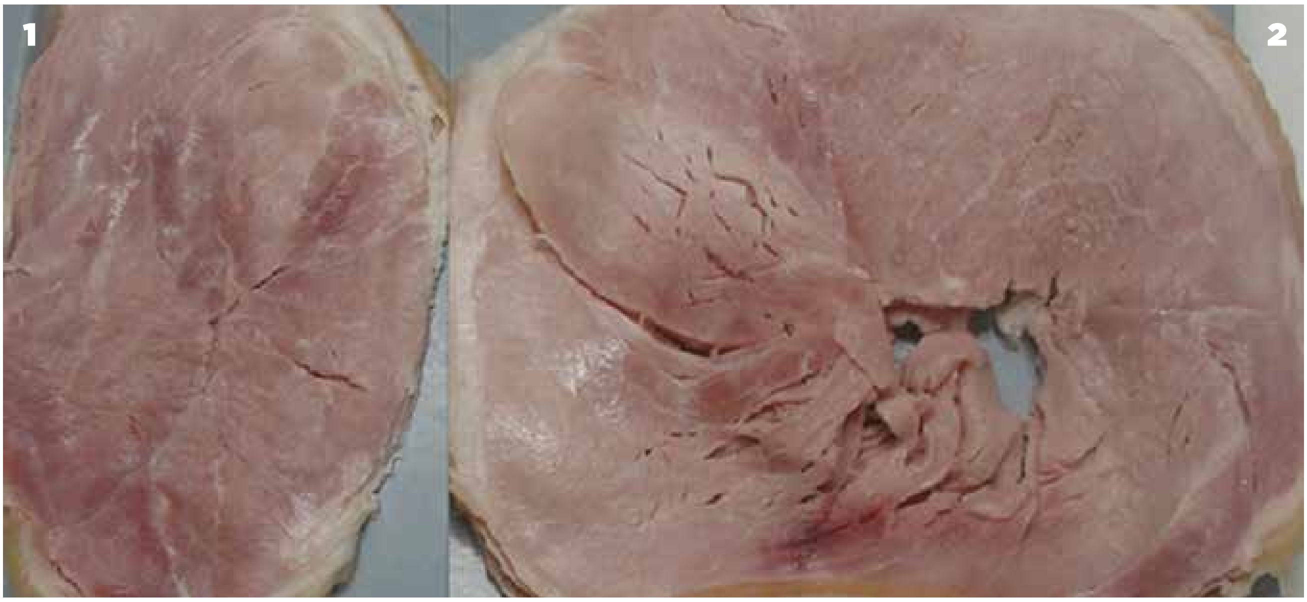

Figure 1. Normal **(1)** and inferior **(2)** quality hams.

Discriminant analysis (SPSS Statistics, IBM, version 20) was performed in order to investigate the use of 1) the conventional methods, 2) the NIR technique, to classify the fresh hams according to the visual score given to the cooked hams after slicing. Concerning the NIR spectral data, factor analysis was performed to reduce the absorbance data at different wavenumbers into fewer number of factors which were used for further discriminant analysis

Results and discussion

After slicing, the hams were visually classified, 39 hams of the original 55 were classed as normal quality hams; 16 hams showed presence of destructured zones and were classified as inferior quality hams. Results of meat quality parameters, measured on the intact and deboned fresh hams, of both normal and inferior classified cooked hams are shown in Table 1.

Table 1. Mean and standard deviation (SD) of the meat quality parameters, measured on the intact and deboned hams, of both normal (n=39) and inferior (n=16) cooked hams

Intact		**Normal**		**Inferior**		
	Muscle	**Mean**	**SD**	**Mean**	**SD**	**P**
pH 30 min p.m.	SM	6.04	0.33	5.69	0.21	<0.001
pH 24h p.m.	SM/AD	5.58	0.14	5.54	0.12	0.340
PQM 24h p.m.	SM/AD	11.38	2.92	10.60	3.04	0.380
Deboned						
pH 24h p.m.	SM/AD	5.57	0.15	5.51	0.19	0.240
pH 24h p.m.	BF	5.68	0.20	5.58	0.12	0.070
PQM 24h p.m.	SM/AD	12.13	2.74	10.63	2.59	0.070
PQM 24h p.m.	BF	12.17	2.70	10.40	2.41	0.030
L* 24h p.m.	SM/AD	55.55	5.29	62.48	3.58	<0.001
L* 24h p.m.	BF	47.88	6.12	54.83	5.92	<0.001
a* 24h p.m.	SM/AD	10.46	1.64	10.56	3.56	0.880
a* 24h p.m.	BF	11.92	2.07	11.64	3.02	0.700
b* 24h p.m.	SM/AD	14.91	5.95	19.19	1.91	<0.01
b* 24h p.m.	BF	13.40	5.95	18.35	1.68	<0.01
Sarcopl. (mg/g)	SM/AD	62.08	8.80	52.49	4.81	<0.001
Myofibr. (mg/g)	SM/AD	10.35	2.36	8.36	0.73	0.002

Cooked hams exhibiting inferior quality had significantly lower pH_{30min} and significantly higher L^*_{24h} and b^*_{24h} values than cooked hams with normal quality. Concerning the technological quality parameters, cooked hams with inferior quality had significantly higher cooking losses than hams with normal quality (Table 2).

Table 2. Mean and standard deviation (SD) of percentage drip loss and cooking loss of normal and inferior cooked hams

%	**Normal**			**Inferior**			P
	n	Mean	SD	n	Mean	SD	
Drip loss*	37	1.05	0.99	10	1.61	1.36	0.150
Cooking loss	39	5.29	1.22	16	6.91	1.14	<0.001

* analyzed on the fresh hams

Both conventional and NIR data were used to classify the fresh hams according to the visual score that was given after slicing. Classification results after cross-validation, based on the measurements carried out 1) on the raw, intact hams and 2) after deboning, for both normal and inferior classified hams are shown in Table 3. Measuring NIR spectra on the BF muscle after deboning resulted in a 87.2% and 62.5% correct classification after cross-validation for normal and inferior hams, respectively, which is approximately equal to the classification obtained by combination of PQM_{24h} + L^*_{24h}, i.e. 89.7% and 62.5% for normal and inferior hams, respectively. Based on sarcoplasmic protein solubility, a 87.2% and 50% correct classification after cross-validation for normal and inferior quality hams, respectively, was achieved. Based on cooking losses, 89.7% and 50% of normal and inferior hams, respectively, could be classified correctly after cross-validation.

Table 3. Percentage correct classification after cross-validation for both normal (n=39) and inferior (n=16) classified hams

Intact	Muscle	Normal	Inferior	Total
pH 30 min p.m.	SM	89.7	43.8	76.4
NIR spectra	BF	87.2	43.8	74.5
Deboned				
PQM 24h p.m. (1)	BF	94.9	0.00	67.3
L* 24h p.m. (2)	SM/AD	87.2	50.0	76.4
L* 24h p.m.	BF	87.2	50.0	76.4
b* 24h p.m.	SM/AD	100	6.30	72.7
b* 24h p.m.	BF	97.4	12.5	72.7
(1) + (2)		89.7	62.5	81.8
Sarcopl. (mg/g)	SM/AD	87.2	50.0	76.4
NIR spectra	BF	87.2	62.5	80.0

The scientific conclusions

The classification result obtained, based on NIR spectral data, is promising. However, further improvement is needed so that more inferior hams could be classified correctly and a more robust classification tool can be achieved.

Future research

Currently, the dataset used, shows an imbalance between normal and inferior quality hams which could possibly have a negative influence on the classification results. Further research should aim to expand the dataset with inferior quality hams. As the signal to noise ratio of the collected NIR spectra is relatively small, spectral processing should be investigated. To test the accuracy of the classification tool a validation in different meat companies is needed.

Acknowledgements

This research was financed by IWT Flanders (Brussels, Belgium), Belpork npo, meat companies and suppliers of NIR equipment. The authors would like to thank the participating slaughterhouses for their assistance.

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Prediction of ultimate pH in beef from NIR spectra collected twenty to forty minutes post mortem

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Value for industry

- The ability to forecast meat quality indicators and attributes developed post-rigor from measurements pre-rigor allows the management of the carcass prior to boning.
- Hot boning is a common practice in the New Zealand meat industry where muscles are harvested from the carcass and packed within hours from slaughter. The muscle cannot be assessed when it reaches rigor since they are already packed.
- Near Infrared Spectroscopy (NIR) allows for fast (< 30 seconds) and a non-invasive measurement. The NIR probe has a pistol based design allowing easy access to the muscle surface, and the whole system is portable.
- Ultimate pH (pH_U) is an indicator of colour, shelf life and tenderness. However, in a hot boning plant it is not feasible to measure pH_U as cuts are already packed when meat reaches pH_U. Thus prediction of pH_U using NIR spectra collected pre-rigor would allow us to predict non-invasively the final quality of the meat cuts.

Background

Following slaughter, a series of biochemical and structural changes take place in the carcass during the conversion of muscle to meat, which will define the final meat quality attributes in the muscles from that carcass. Several factors during pre-slaughter (i.e. natural biological animal-to-animal variation, pre-slaughter handling) and processing such as variations in the electrical inputs, stimulation, temperature will influence the conversion of muscle to meat (Hwang *et al*., 2003). Not all these factors can be controlled during the process and inconsistencies in the carcass meat quality can occur. A strategy to deal with these inconsistencies is to identify abnormal carcasses early in the process and manage them towards a desire set of attributes. To be successful, this strategy requires the carcass to be evaluated within a shorter time frame that allows it to be managed. In a hot boning situation, this time frame can vary from 20 minutes to 1 hour from slaughter in the same plant, depending on chain speed.

Why work is needed

The pH in meat 48 hours post slaughter (ultimate pH, pH_u) is an indicator for meat quality attributes (colour, tenderness, etc.,) and shelf life (Braggins *et al*., 1996; Dransfield *et al*., 1981; Gill and Newton., 1981; Purchas and Aungsupakorn., 1993; Silva *et al*., 1999; Viljoen *et al*., 2002; Watanabe *et al*.,1996). Studies carried out in New Zealand have revealed the incidence of carcass with pH_u higher than 5.8 to be 15% for prime animal and 50% in bulls (Graafhuis and Devine, 1994; Wiklund *et al*.,2009; Young *et al*, 2004).

Meat with high (≥ 5.8) pH_u presents undesirable attributes, such as dark cutting, shorter shelf life and inconsistent tenderness (Braggins *et al*, 1996; Dransfield *et al*, 1981; Gill and Newton, 1981; Purchas and Aungsupakorn, 1993; Silva *et al*, 1999; Viljoen *et al*, 2002; Watanabe *et al*, 1996).

In cuts from a hot boning plant pH_u cannot be measured as when the meat reaches its pH_u, cuts are already deboned, vacuum-packed and in storage. While there are invasive methods to forecast the meat pH_u from samples collected pre rigor (Young *et al.*, 2004), a fast and non-invasive system is desirable and NIR is one of the potential candidate for it.

The methods used

The NIR spectra were collected (between 20 and 40 minutes from slaughter) from the carcasses (cows *n* = 86, bulls *n* = 170, steers *n* = 363 and heifers *n* = 38) in a commercial abattoir during their normal processing routine. Collection was carried out in 6 days distributed in three different weeks (Reis and Rosenvold, 2013). NIR spectra were collected on a transverse section of the *M. longissimus dorsi* from the surface exposed at the quartering cut (between the 11th and 12th rib). Spectra were collected using a LabSpec 5000 (ASD Inc., USA) with a customized reflectance probe that has similar internal design as the standard high intensity probe from ASD. Spectra were collected in the spectral range from 350-2500 nm, with wavelength accuracy of ± 1 nm (Reis and Rosenvold, 2013). A slice of the scanned area in the muscle (10 cm think) was excised in the boning room, vacuum packaged and chilled for 48 hours under the normal plant chilling conditions. pH_u was measured 48 hours later using a calibrated pH probe (Testo 205 pH meter; Lenzkirch, Germany) placed in three different locations and averaged. A subset of samples were aged between 9 and 10 days under normal ageing conditions of the plant, then frozen and sent to AgResearch for further analysis. The samples were cooked from the frozen state in a 100°C water bath. When an internal temperature of 75°C (as measured by a thermocouple) was reached each sample was immediately placed in ice-water slurry. Once cooled, ten sub-samples with a 10 mm x 10 mm cross section were dissected and sheared using a MIRINZ tenderometer. The mean shear force (KgF) over the 10 sub-samples gave the shear force for each sample.

Partial least square models (PLS) (Wold *et al*.,2001) were fitted to predict pH_u. The original data were split into a calibration and validation data sets (~ 50% each), with carcasses distributed between these sets to cover the full variation of the original data set with respect to pH_u, animal categories (bulls, steers, heifers and cows) and days of data collection (Reis and Rosenvold, 2013). Three different models were fitted: A model including the spectra from all four categories of animals (All-animals); a model including the spectra from heifers, cows and steers but excluding the spectra from bulls (Non-bull model); and finally a model including only the spectra from bulls (Bull model). The number of latent variables for each of these three models were chosen using cross validation with n segments of samples randomly selected (non-bulls: *n* = 20; bulls: *n* = 9; all animals: *n* = 10). Spectra were pre-processed using extended multiplicative scatter correction (EMSC) and orthogonal signal correction (OSC). The R^2 and root mean square error (RMSE) were used to assess the models performance. Data analysis was carried using R v.2.9.1(R-Development, 2009), Unscrambler X (v. 10.2, CAMO software AS), PLS-Toolbox (v.4.1.1 Eigenvector Research Inc.) running on Matlab® 7.4. (The MathWorks, Inc. (Reis and Rosenvold, 2013).

The results obtained

The best models produced an R^2 for internal cross validation of 0.50 (RMSE: 0.26), 0.41 (RMSE: 0.24) and 0.43 (RMSE: 0.33) for the All-animals-model, Non-bulls model and Bulls-model, respectively. The predictions of these models were used to classify carcass as high pH_u ($pH_u \geq 5.8$) or normal ($pH_u < 5.8$). The best models were those fitted specifically for each category, Non-bull and Bull models, although predictions for Bulls are not as good as for the Non-bulls model (Reis and Rosenvold, 2013). The Non-bulls model correctly classified 90% of the high pH_u carcasses and 89% of the normal pH_u carcasses. The Bulls model correctly classified 94% of the high pH_u carcasses and but only 65% of the normal pH_u carcasses.

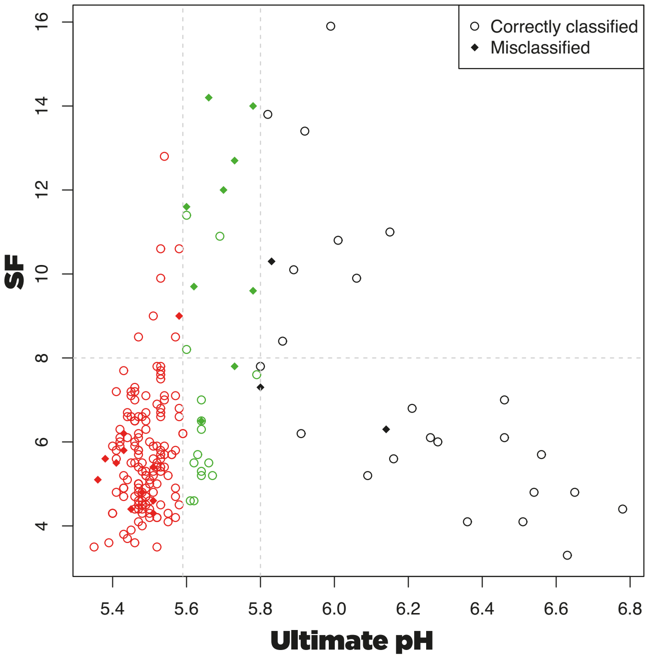

Figure 1. Distribution of shear force values (KgF) according ultimate pH for Non-bulls validation date set (n=177).

Figure 1 present the distribution of shear force values according measured pH_u for the samples in the validation data set for the Non-bulls model. It has a bell shape curve were the high and low ultimate pH ends present lower value of shear force, while the center of the curve presents a wider distribution of shear force values. The analysis of this distribution of shear force values (Figure 1) revealed that the highest rate of misclassified samples according pH_u lies between 5.6 and 5.8 where the shear force is the most variable. These missclassified samples in general have a higher value of shear force, compared to the others correctly classified samples in the same category. Indeed, the Non-bull model discarded samples with high shear force values, when misclassified the samples with pH_u between 5.6 and 5.8 as being high pH_u. For this particular case, samples classified correctly as normal pH are more likely to present shear force value below 8 KgF.

The scientific conclusions

Carcasses could be classified as pH_u above or below the threshold of 5.8 from spectra collected up to one hour post slaughter.

There is a potential to use NIR based system to predict pH_u.

The classification could also be used as proxy for shear force values, which needs to be verified.

The next steps

To evaluate whether seasonal variations influence the performance of the models.

To investigate further the relationship among NIR, pH_u and the shear force.

Acknowledgements

The authors acknowledge Silver Fern Farms for funding the project and providing the sites for data collection. The New Zealand Ministry for Primary Industries contributed through the Primary Growth Partnership – PGP programme. Kevin Taukiri for assisting with NIR data collection. Grant Pearson for reviewing the manuscript. Daniel Lousich and Silver Fern Farms staff provided valuable support for the trials at the plant.

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Project overview – large scale assessment of the utility of spectral technologies in the beef sector

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Value for industry

- Routine collection of eating quality data could allow quality information to be utilised by the value-chain for monitoring and refining production and processing to optimise meat product quality.
- Non-destructive assessment of some beef meat quality parameters (e.g. slice shear force and pH) applied online in the processing plant could be used to improve consistency of meat and meat products.
- An objective eating quality measurement would enable the value chain to improve consistency and possibly seek a premium based on intrinsic meat eating quality parameter(s).

Background

Matching supply with customer demand is a challenge for the meat value chain (Zokaei & Simons, 2006). At a macro level, this is partly attributable to social, economic, political and climatic factors, but also because customer preferences of meat attributes are subject to change (Tonsor, Mintert, & Schroeder, 2010) and vary by country and region (Sañudo *et al*., 1998). At an individual supplier level, a key contributing factor is that customer preferences are not effectively relayed back along the value-chain and are not therefore reflected in market signals.

One reason for the poor communication of customer preferences is that these preferences are not accurately reflected in the various carcass evaluation systems used around the world (Polkinghorne & Thompson, 2010). The EUROP carcass evaluation system operated in the European Union is no exception because it focuses on carcass conformation and fatness parameters (Fisher, 2007) and makes no direct assessment of meat eating quality. More effective value-based marketing (VBM) systems have been advocated as a potential solution to this problem (Cross and Whittaker, 1992; Johnson and Ward, 2006; Polkinghorne *et al*. 2008) yet few meat value chains have actually adopted a VBM system at an industrial level.

The main value attributes of a carcass are currently the saleable meat yield and the eating quality of the meat. Therefore saleable meat yield and eating quality need to be measurable and marketable. Saleable meat yield can be predicted using video image analysis (VIA) systems (Craigie *et al*. 2012), but there is no direct assessment of meat eating quality at present - largely due to the lack of a suitable measurement technique.

Near-Infrared spectroscopy (NIRS) and Hyperspectral imaging (HSI) techniques have shown promise within individual plants, but the feasibility of developing models within and between processing sites has not been investigated.

Why the work is needed

The present objectives are to:

1. **To quantify the variation in meat product quality from the *M. longissimus thoracis* across the Scottish beef industry and feed this information back to meat processors.**
2. **To assess NIR and HSI for predicting slice shear force and ultimate pH under a range of processing conditions using new, novel analytical approaches.**
3. **To establish the robustness of the imaging techniques within the Scottish beef industry.**

The methods used

Sampling was undertaken in five beef processing facilities across Scotland. Over two days, >200 slices of *M. longissimus thoracis* ≥25 mm thick were recovered from the 11th rib position of the strip-loin at 48-hrs post mortem. Steaks included in the analysis were chosen from the half-carcasses available on the trial day and were selected irrespective of gender, conformation, fatness, weight or maturity. Where possible, steaks were selected from different carcasses to maximise variation, but this was only possible in 3 abattoirs where carcass ID was available from kill tags. In plants where tags were unavailable, it is possible that a steak was recovered from left and right sides of the same carcass.

After recovery, steaks were allocated a number, and were allowed to bloom for at least two minutes before imaging with :

1. A hyperspectral imaging system [HSI] (400 -800 nm, Specim V8E attached to a Andor Luca CCD camera, mounted above a translation stage and illuminated by a 250 Watt halogen flood lamp) (Figure 1).
2. An NIR spectrophotometer [NIRS] (ASD Labspec Pro (350-2500 nm) fitted with a internally-illuminated probe with a 68 mm ø active scanning area (Figure 2).

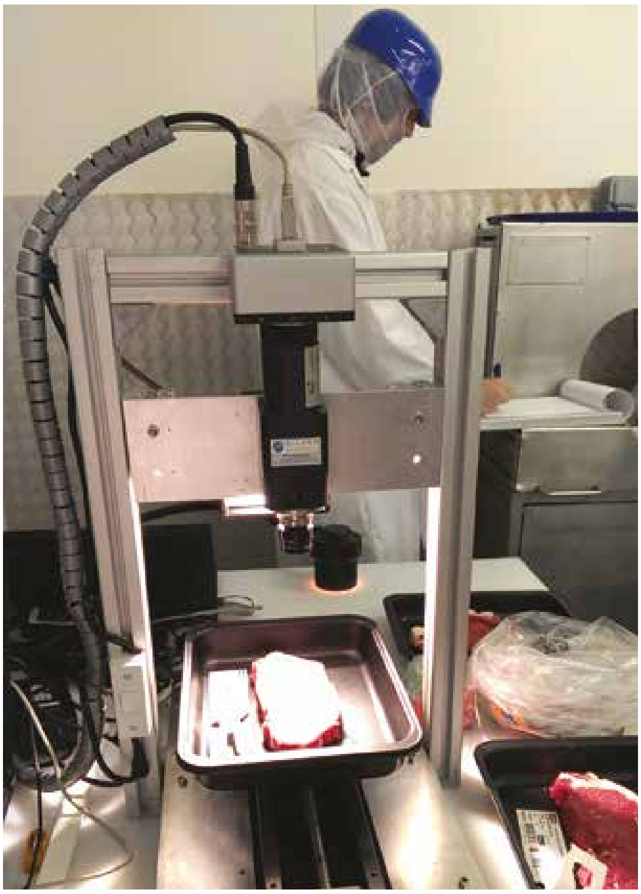


Figure 1. The HSI system showing an *M. longissimus thoracis* steak being imaged.

Figure 2. The NIRS system showing the measuring head applied to an excised *M. longissimus thoracis* steak.

For each steak, a single HSI image and 10 replicate NIRS scans were captured. After imaging, each steak was divided into dorsal and lateral halves, labelled and vacuum packaged. The dorsal halves were boxed and aged for an additional 12 days at ~ 1 °C before freezing and the lateral halves were boxed and aged for 5 days before freezing to give 7 and 14d aging respectively. Each box of samples contained a temperature data logger to enable the aging duration and temperature to be verified. Samples from each processing plant were defrosted at ambient temperature for 24 hours in lots of approximately 100, such that 7 and 14d aged samples from the same carcasses were defrosted together. Ultimate pH was measured on each half steak using a calibrated Hanna meat pH meter (HI 99163) without the knife blade attached. Two replicate measures were taken and averaged to give the final reading. Following pH measurement, steaks were cooked to a core temperature of 71°C on a George Foreman clam-shell grill. A slice shear force measure (Shackelford *et al*. 1999) was taken on each half steak with a Tenderscot tenderometer (Pentland Precision Engineers, Loanhead, Midlothian).

The force data from the Tenderscot (Figure 3) were captured directly into Microsoft Excel® 2003 using a macro, and the steak ID was assigned to the force deformation curve. The peak force (PF) was extracted from each force deformation curve and the work done (WD – the area under the force deformation curve) was calculated after converting any negative force values to zero. Negative force values are captured by the macro during the return phase of the blade after shearing. In summary, the following instrumental meat quality parameters were measured on each steak: pH7, pH14, PF7, PF14, WD7 and WD14.

Figure 3. The Slice Shear Force (SSF) test.

The results obtained

As the experiment is on-going, only results pertaining to the first objective will be presented in the current report. The texture and pH of >1000 beef *M. longissimus thoracis* steaks from five Scottish beef processing plants (circa 200 steaks per plant) have been sampled to date. As of 2012, these five processing plants account for around 60% of the Scottish beef processing capacity. The use of a consistent methodology across a number of plants allows overall profiles of beef texture and pH to be generated. This reference meat quality information is being used to develop algorithms that can determine the PF, WD and pH of beef *M. longissimus thoracis* from NIRS and HSI images. Some preliminary results on predictive ability of NIR and HSI were presented by J. Zabalza *et al*. and T. Qiao *et al*. at the FAIM II conference. This profile is a baseline measure of beef product quality for the Scottish beef processors which the industry can use for benchmarking purposes. The distributions for PF7 and PF14 (Figure 4), WD7 and WD14 (Figure 5) and pH7 and pH14 (Figure 6) are shown below. It can be seen from the plots showing the distribution of the meat quality traits, that PF and pH parameters are highly skewed. It is also noteworthy that the PF7 and PF14 curves are almost identical (Figure 4) which suggests that either the additional 7d aging has little effect on the slice shear force, or that there is differences within the steak. The sampling position in the steak (dorsal) and the aging time (14d) are confounded in the present experiment, further work is needed to establish whether or not a positional effect is masking the aging effect. An increase in pH by approximately 0.05 units was also seen with the additional 7 days aging (Figure 6).

Figure 4. The percentage distribution of peak shear force values for beef *M. Longissimus thoracis* steaks when cooked to 71°C after aging for 7 and 14 days.

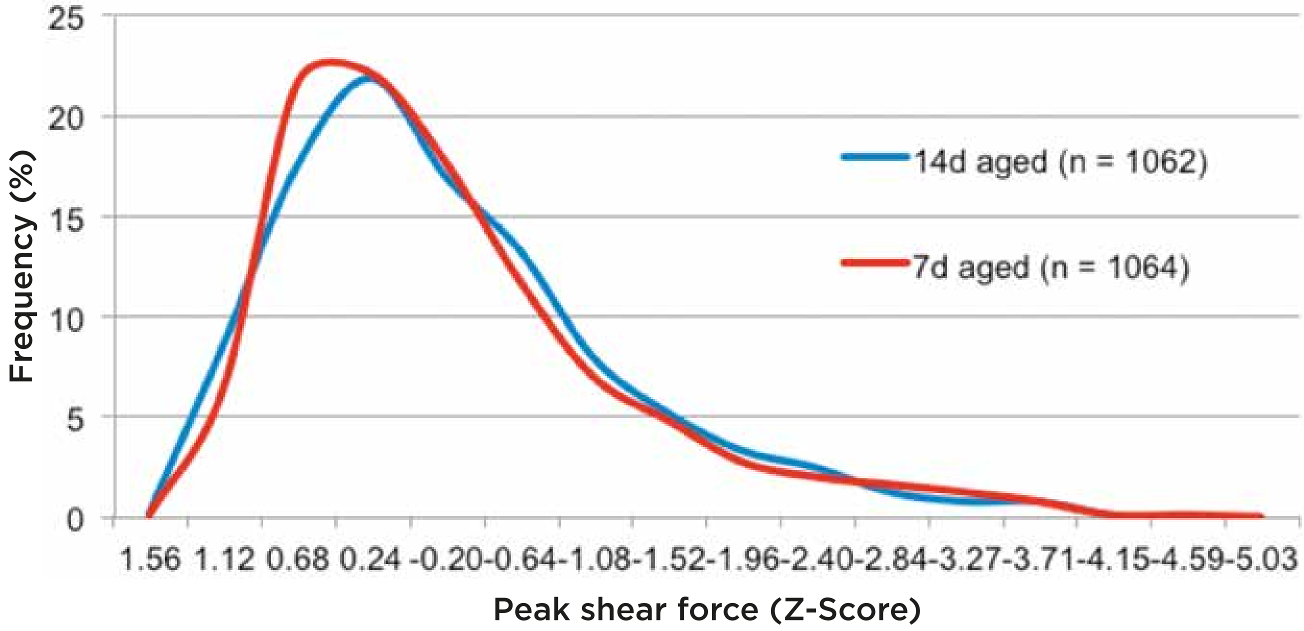
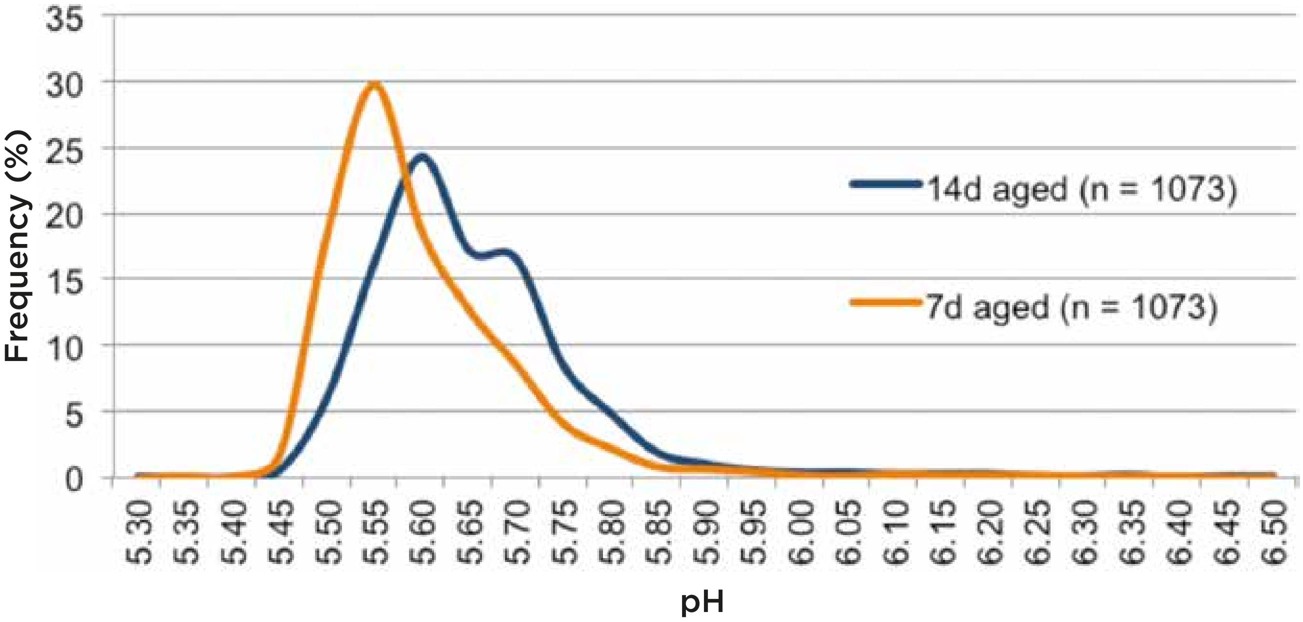


Figure 5. The percentage distribution of work done (area under the force deformation curve) for beef *M. Longissimus thoracis* steaks when cooked to 71°C after aging for 7 and 14 days.

Figure 6. The percentage distribution of pH for *M. longissimus thoracis* steaks after aging for 7 and 14 days.



Variation in meat quality parameters between plants relative to the average are shown for PF7 (Figure 7) and PF14 (Figure 8), as well as WD7 and WD14 (Figure 9) and pH7 and pH14 (Figure 10). As can be seen from the plots, there are clear differences between processing plants. Quantifying the source of the variation is not a primary objective of this work, but provided that the animal ID is available for each steak, a breakdown of data for each processor from samples collected at their premises is possible. The proportions of steers, heifers, young bulls and cows in the samples are shown in table 1.

Carcass ID tags contain supplier, genotype, gender, weight and classification data, therefore it is possible to summarise the meat quality data on the basis of those parameters. Although the data are currently only available for around 200 cattle per processor, presenting a break-down of data in this way could quickly show trends, and enable processors to identify sources of variation in meat product quality. However, the ability to collect meat quality data in a non-destructive way currently is limited.

Figure 7. Differences in *M. longissimus thoracis* peak shear force between plants (expressed as a percentage relative to the overall average) after aging for 7 days.

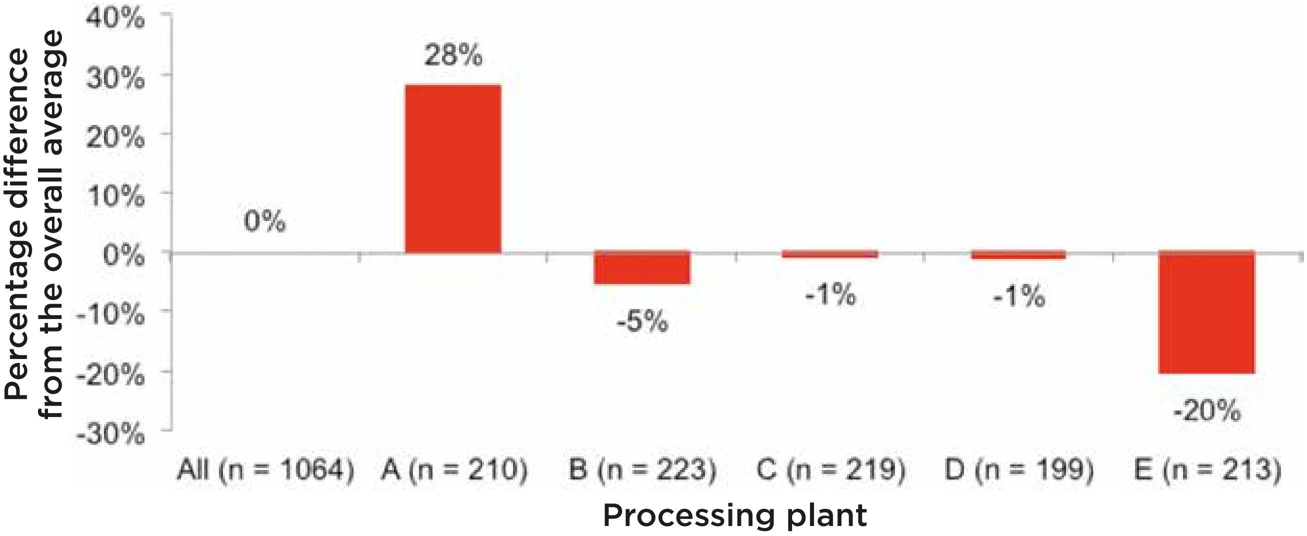


Figure 8. Differences in *M. longissimus thoracis* peak shear force between plants (expressed as a percentage relative to the overall average) after aging for 14 days.

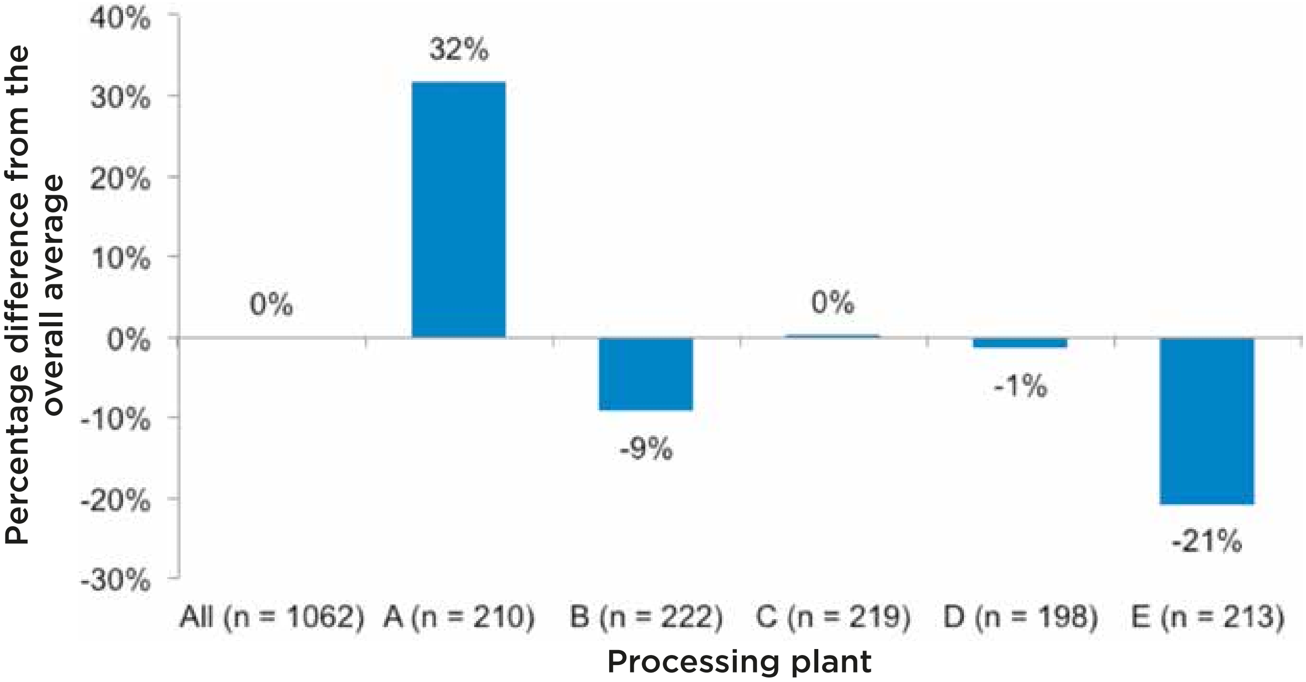


Table 1. Gender of cattle included in the sample at each plant

Plant	Steers	Young Bulls	Heifers	Cows
A	35.27%	50.72%	13.53%	0.48%
B	55.41%	21.62%	22.97%	0.00%
C	63.89%	0.00%	36.11%	0.00%
D*	47.27%	0.00%	44.55%	8.18%
E*	56.10%	0.00%	43.90%	0.00%

*percentages are indicative only, as kill tags were only partially available

Figure 9. Plant differences in *M. longissimus thoracis* work done (area under the force deformation curve) after cooking to 71°C from meat aged for 7 and 14 days.

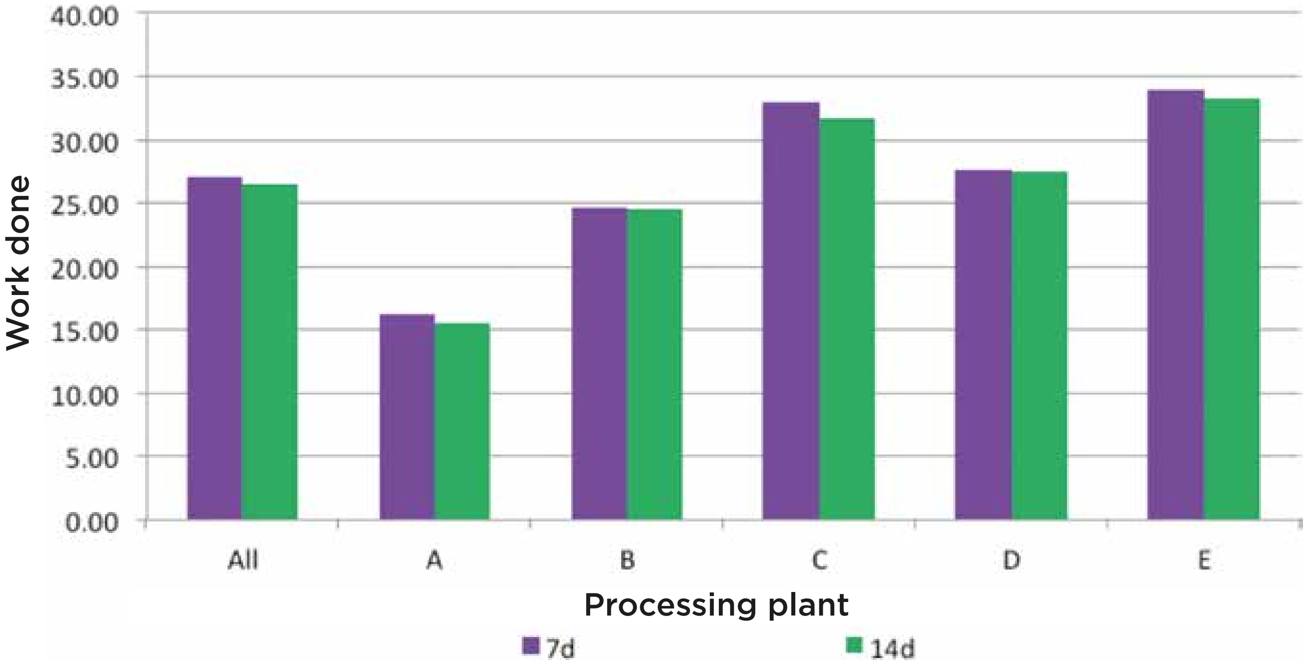
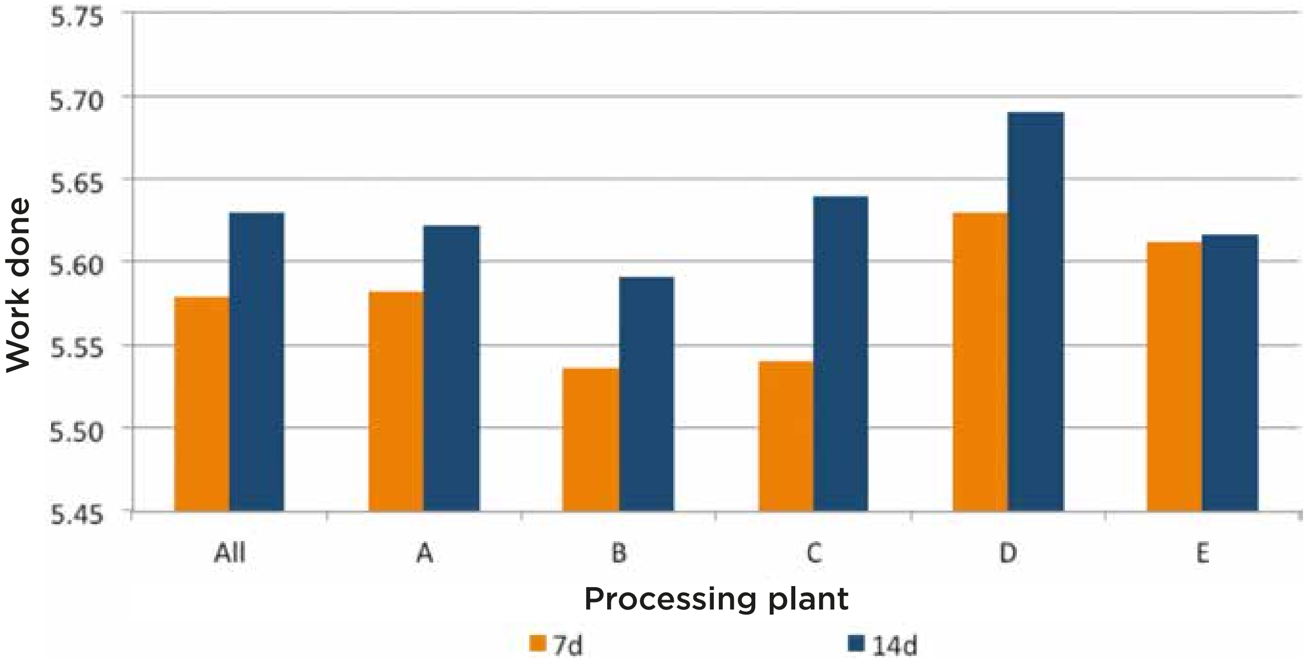


Figure 10. Plant differences in *M. longissimus thoracis* pH after aging for 7 and 14 days.



The scientific conclusions

This work presents the texture and pH of >1000 beef *M. longissimus thoracis* steaks from five Scottish beef processing plants (circa 200 steaks per plant) sampled to date. From the data, it is clear that there is variation in meat texture and pH between processing plants.

This research has already shown that measuring beef meat quality and relating the quality data back to animal ID provides information which can be used to identify some sources of variation in meat product quality by the value chain.

Further research is needed to establish whether the position (dorsal or lateral) within the *M. longissimus thoracis* affects the texture.

Development of NIRS and HSI techniques to measure meat quality on-line will enable processing plants to quantify meat quality on a routine basis, and will enable the sources of variation to be identified and controlled in the future.

Measurement of meat eating quality information has the potential to add value through improved product consistency for quality attributes, and raises the possibility for a guaranteed premium range that can be marked accordingly.

Acknowledgements

The authors wish to acknowledge meat processor staff and Miss Dominique Daly for assistance in data collection.

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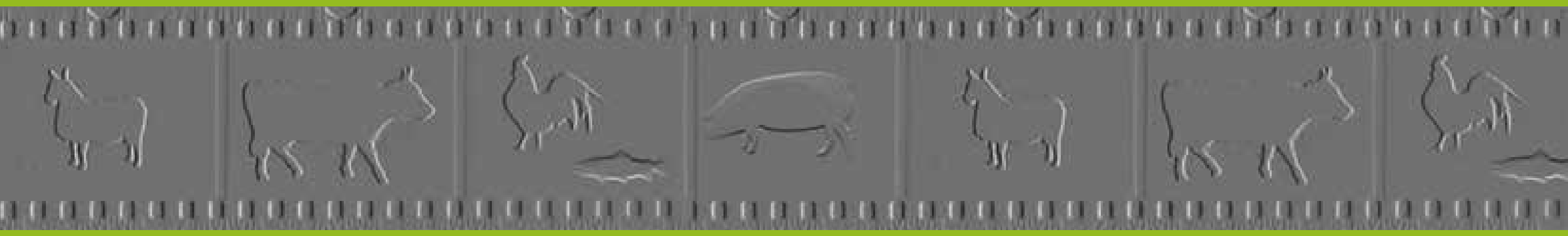

Parametric representation of lamb carcasses based on computed tomography (CT) images

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Value for Industry

- This project aims to build a robust yield predictive model based on an accurate description of a carcass built from CT scans which can be morphed to represent a particular carcass using any available information on the live animal and carcass.
- Independence from any particular objective measurement technology and specific primal and retail cuts will allow adaption as new measurement technologies emerge and innovative meat cuts are considered.
- The model will be used to generate predictive equations that will be embedded in meat grading systems with a target analysis rate of 30 carcasses per minute. This will permit extension to yield predictions for cattle and pigs.

Background

With an annual turnover of €1.2b, Silver Fern Farms processes 30% of all cattle, sheep, lambs & deer in New Zealand (Buchanan, 2012). Silver Fern Farms uses a number of objective measurement technologies in its production lines. In 2010, Silver Fern Farms deployed the world's first X-ray analysis system for lamb, now operational across all of the company's sheep and lamb plants. The X-ray data is being used for primal weight predictions on all sites and to make cutting decisions form automated primal separation equipment on two sites (Buchanan, 2012).The X-Ray system is being further developed with DEXA hardware for on-line meat/fat/bone analysis.

The Auckland Bioengineering Institute (ABI) is a cross-faculty research institute within the University of Auckland. It deals with the application of the mathematical and engineering sciences to biology generally and human physiology in particular. ABI has decades of research experience in subject-specific geometric and biomechanic modelling, and has its own modelling software (e.g., CMISS, CMGUI) (Bradley *et al*, 1997).

In 2013, Silver Fern Farms and ABI started an innovative carcass modelling project in which the major lamb carcass muscles and bones are described mathematically into a generic 3D computer model. The ultimate aim is to apply the model to predict various features of individual muscles and retail cuts from measurements of carcass variables on the processing chain at a rate of up to 30 carcasses per minute. The project has been subdivided into four stages spanning two years development. The first milestone was completed recently.

Methods

A: CT imaging

CT scanning was performed for five carcasses (weight 19.76 ± 2.8 Kg) at AgResearch, Invermay, New Zealand. The spatial resolution was 0.765mm x 0.765mm x 5mm (width x height x depth). One carcass was chosen as the reference subject due to its typical anatomic features (e.g., 13 ribs), so that a generic carcass model could be built.

B: Generic carcass model

The pipeline of generating a generic carcass model is shown in Figure 1. In brief:

- The CT images were loaded into a visualisation and imaging software CMGUI (http://www.cmiss.org/cmgui);
- A 3D data cloud delineating various carcass tissues (muscle, fat, skin) was created from the image slices;
- The data points representing different tissues were grouped in a systematic manner;
- Based on the data cloud, 2D cubic Hermite meshes which had C^1 continuity were fitted onto the data cloud yielding the carcass geometry.

C: Simulation of carcass cuts

Primal and retail carcass cuts can be simulated from the generated 3D model using graphic techniques. For example, an arbitrary carcass cut is shown in Figure 2(a). This is realised by using spectrums along the X, Y, Z directions which yield the carcass cut ranges. By adjusting the spectrums we are able to simulate actual or imaginary cuts. Note that the digitised muscles and fat tissue are also shown at the cross section.

D. Generic carcass mesh morphing

The core of the mesh morphing algorithm is to determine the appropriate transformation matrix, which is evaluated by fitting a set of landmark points (fiducial points) from the reference carcass to their corresponding target points on individual carcasses. The landmark points are placed inside a 'host' finite element mesh whereby their local coordinates are computed. After the fitting procedure the host mesh is transformed so that coordinates of the target points remain the same as in the deformed host mesh. The 'slave' mesh, i.e., the generic carcass mesh is then morphed to individual carcasses. The morphed models are then compared with their respective CT images for validation. The key components of this 'host-mesh-fitting' algorithm, including the host mesh and the landmark/target points, are shown in Figure 2(b). At this stage we are using a cylinder-shaped 2D mesh (resolution 14x12, i.e. total 168 node points) to match the landmark points and target points.

Figure 1. The pipeline for creating a generic carcass mesh.

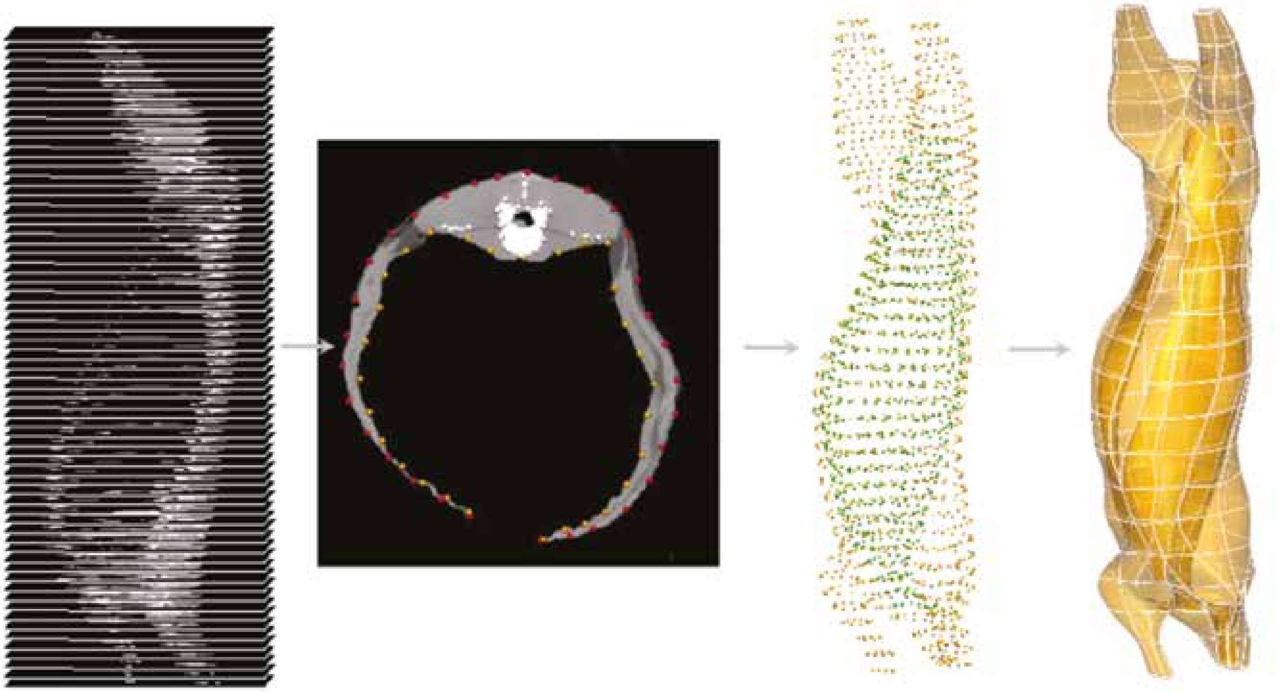
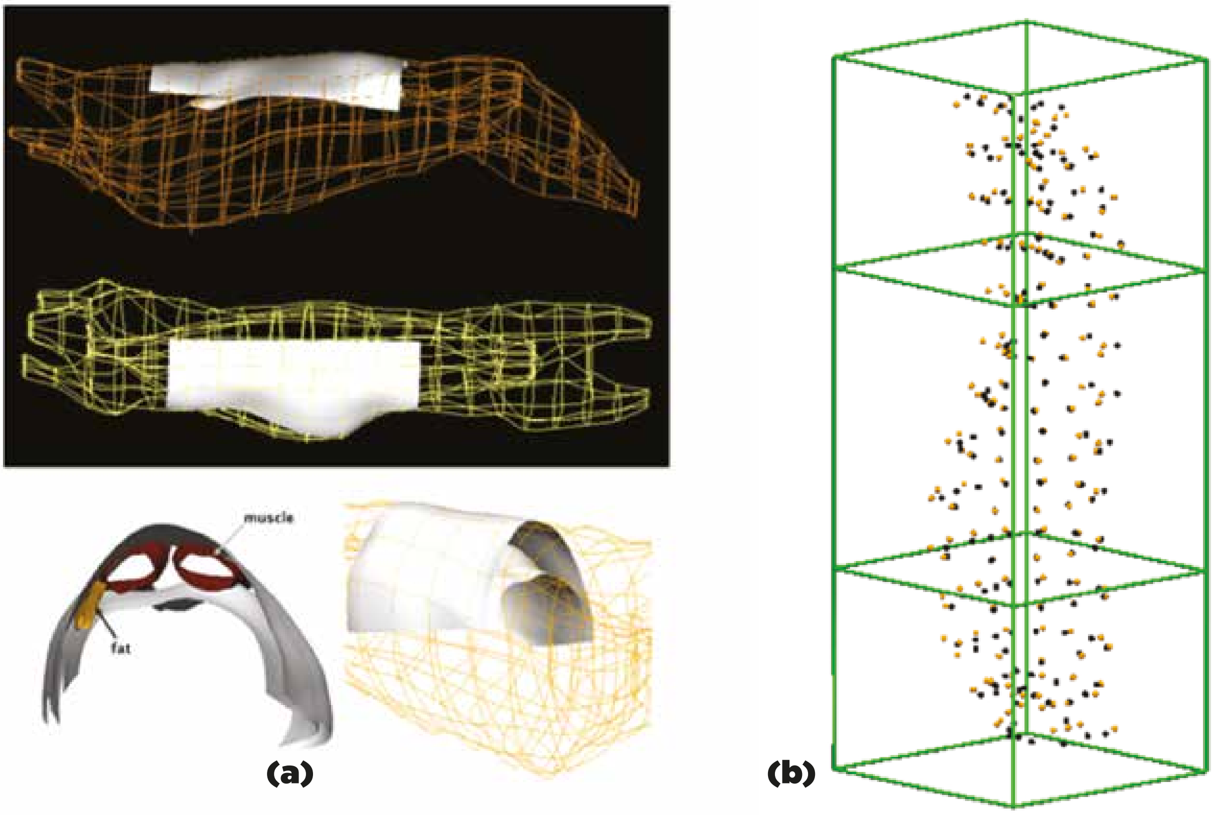


Figure 2. **(a)** An arbitrary carcass cut; **(b)** Key components in the morphing algorithm: 168 landmark points (in gold colour) and target points (black) are placed inside the host mesh (green).



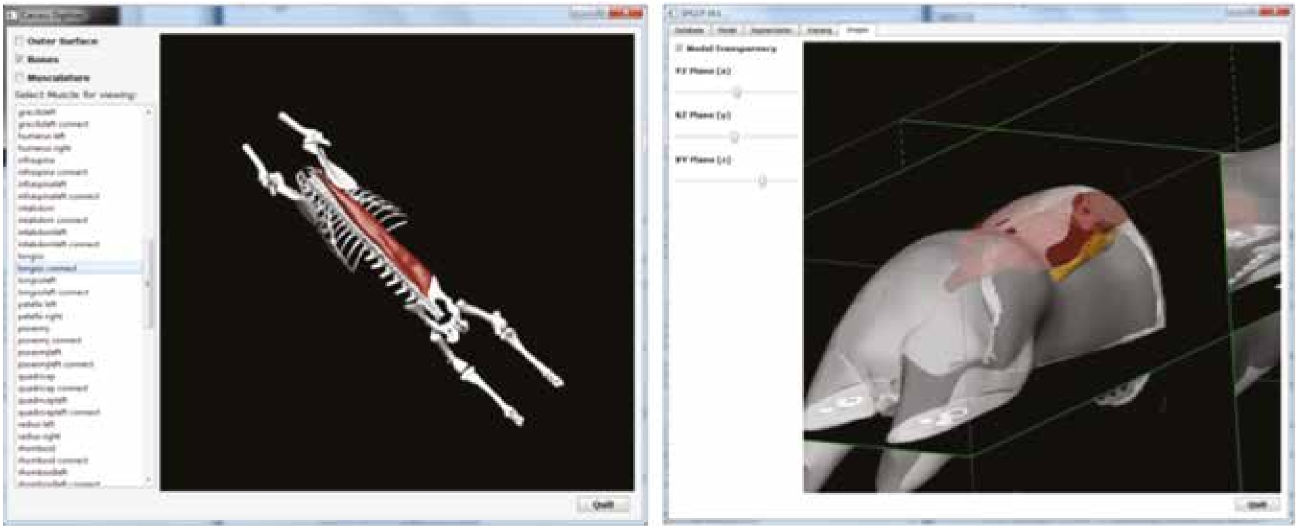
Results

We have run the whole cycle of the carcass morphing pipeline, from CT image digitisation to model validation. The main outcomes include:

- The generic carcass mesh (i.e., that shown in Figure 1) was created, where the internal surface mesh was stitched seamlessly with the external surface mesh;
- Carcass cuts were simulated (e.g., illustrated in Figure 2(a);
- The generic mesh was morphed to four other carcasses. High accuracy was achieved at torso surface, where the landmark/target points were located.

The first version of the modelling software (V0.1) has been developed, built on a 3D rendering engine PyZinc developed by ABI. The software was written in the Python language. Two snapshots of its user interface are shown in Figure 3. More features, e.g., query to image database, are being developed for the software.

Figure 3. Lamb carcass modelling Software (V0.1). Two snapshots of the software are shown here, highlighting the musculature/skeleton module and the CT carcass digitisation module.



The scientific conclusions & next steps

Mathematical models of lamb carcasses and their major tissues have many potential applications. An example of carcass model morphing has been shown in the above sections. In the latter stage of the project, we will adopt a broader modelling approach to make a best guess of carcass proportions and retail cut yields from the data available in the processing environment. This could include gender, age, weight and skeleton dimensions. Future work will establish the accuracy of predictions depending on these available parameters.

Acknowledgements

We thank Wendy Bain of AgResearch, NZ for providing the CT images.

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Gabor-filter based automated removal of troughs from CT images

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Value for Industry

- The proposed method enables the segmentation and removal of troughs from CT images.
- The method is fast, template-free and independent from the shape of the cross-section of the trough until it does not vary heavily along the axial axis.

Background

The resolution of CT imaging is finite, the partial volume effect does not allow the proper reconstruction of fine details and the troughs used to hold the animals during the scanning are usually made of various materials (mostly different plastics). Accordingly, the images of troughs represent a wide range of intensities (proportionally to densities) and deteriorate the automated, intensity based evaluation of the images. The manual masking or semi-automatic segmentation of the trough is an easy task; however, the automated processing of a large number of images requires the automated and precise detection and segmentation of the voxels of the trough in the volumetric representation.

Why work is needed

There are several extensively researched areas of digital image processing, like face and facial gesture recognition, the understanding of satellite images, the recognition of human silhouettes on video streams, etc. Unfortunately, the solutions developed for the popular problems of image processing are far not general: the commonly used approaches always have to be adopted to the different types of images and tasks. In most of the real applications the image processing methods are assembled from panels like contrast enhancement, feature extraction, statistical model fitting and classification. The characteristics and problems of farm animal imaging tend to be rather unique, usually very different within farm animal imaging applications and also different to those problems encountered in the extensively researched area of medical imaging. The common point is that there is usually a trough to hold the living animals or body parts during the scanning. The large amount of image data, the variety of image contents and the requirements for automated processing makes the development of general and automated trough detection and removal algorithms necessary.

The methods used

We have made some mild assumptions about the shape of the troughs used in farm animal imaging. Particularly, the troughs are supposed to be connected objects (some leaks are allowed) and the cross sections of the troughs are supposed to vary slowly and smoothly along the axial axis. The material and the shape of the cross section of the trough can be arbitrary.

The proposed method consists of four main steps: the application of Gabor-filters to enhance the contrast of linear line segments; the thresholding of the filtered image to get a binary segmentation; axial morphology to remove artifacts from the segmentation and the selection of the largest connected component to get the final volumetric model of the trough. In the rest of the section these steps are detailed.

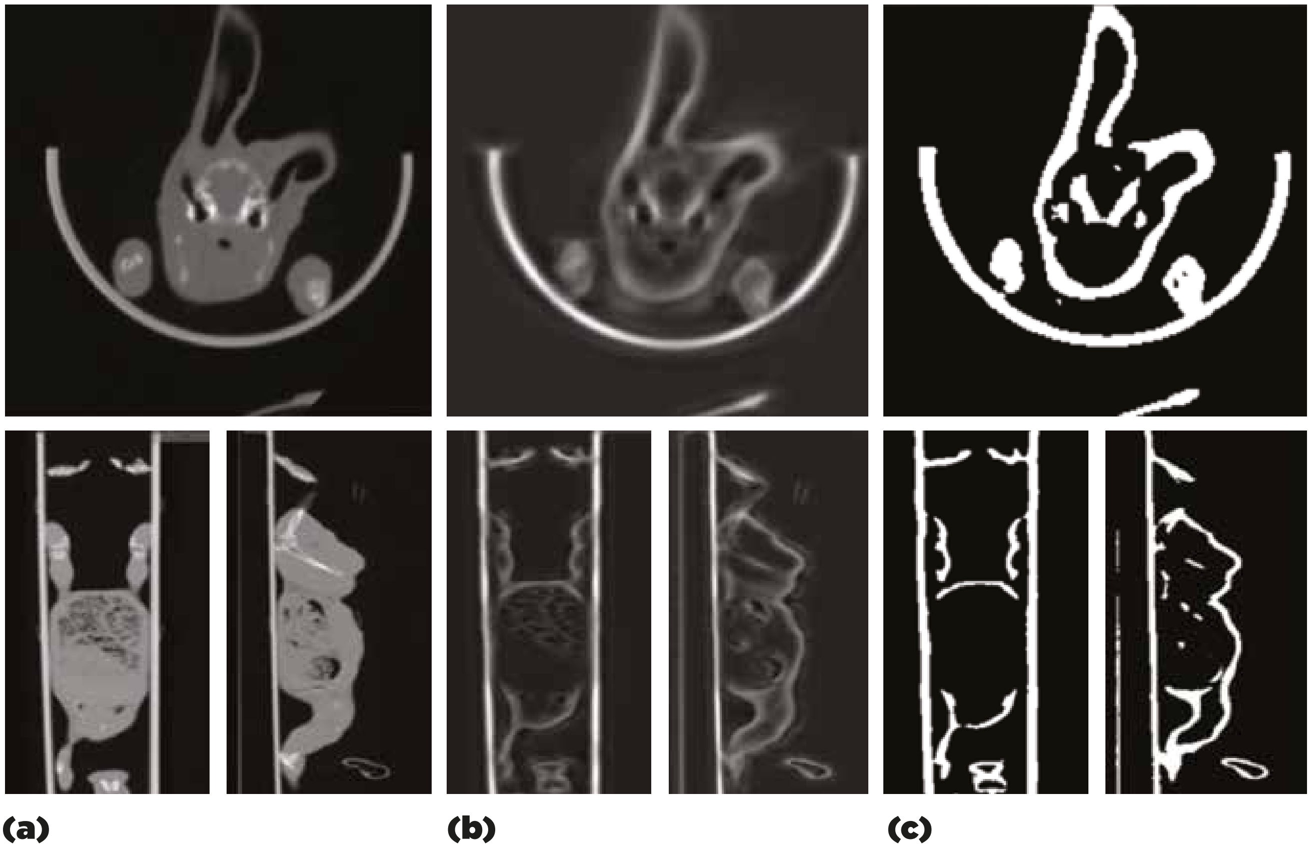


Figure 1. The axial, coronal and sagittal cross sections of the original image **(a)**; the filtered image **(b)** and the thresholded image **(c)**.

Gabor-filters[1] (Feichtinger *et. al.*, 1998) are linear convolutional filters enhancing the contrast of line segments on digital images. Formally, Gabor-filters are defined as the composition of a Gaussian-filter and a complex wave. In practice, we utilize only its real part:

$$G(x,y;\lambda,\theta,\psi,\sigma,\gamma) = e^{-\frac{(x\cos\theta+y\sin\theta)^2+(-x\sin\theta+y\cos\theta)^2\gamma^2}{2\sigma^2}}$$

$$\cos\left(2\pi\frac{x\cos\theta+y\sin\theta}{\lambda}+\psi\right).$$

In this widely used formulation λ denotes the wavelength of the filter, that is approximately twice the width of the line segments one wants to enhance; θ is the angle of the line segments to enhance; ψ is the phase offset of the filter, particularly, $\psi=0$ and $\psi=\pi$ are related to the enhancement of bright and dark line segments, respectively; σ is the width at half maximum of the exponential part, related to the spatial extents of the filter; and γ denotes the aspect ratio of the filter, that is, specifies the ratio of the width and length of the line segment the filter represents.

[1] Proposed by and named after the Hungarian physicist Dénes Gábor, better known as the Nobel-prize awarded inventor of holography. Gabor-filters have been shown to enhance similar features as the visual cortex of mammal brains.

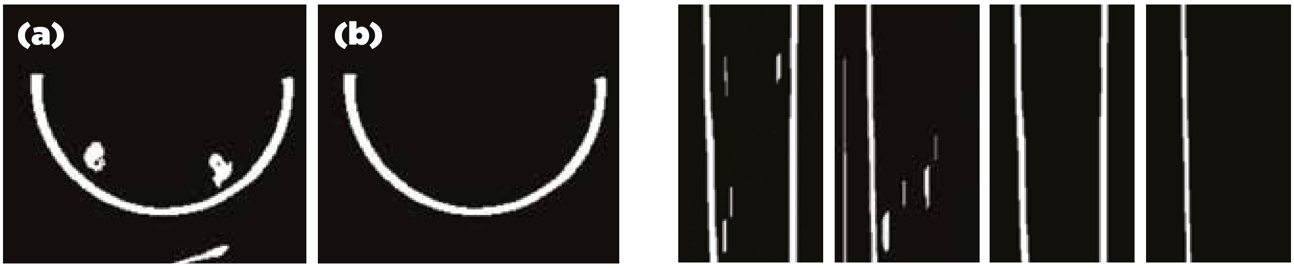
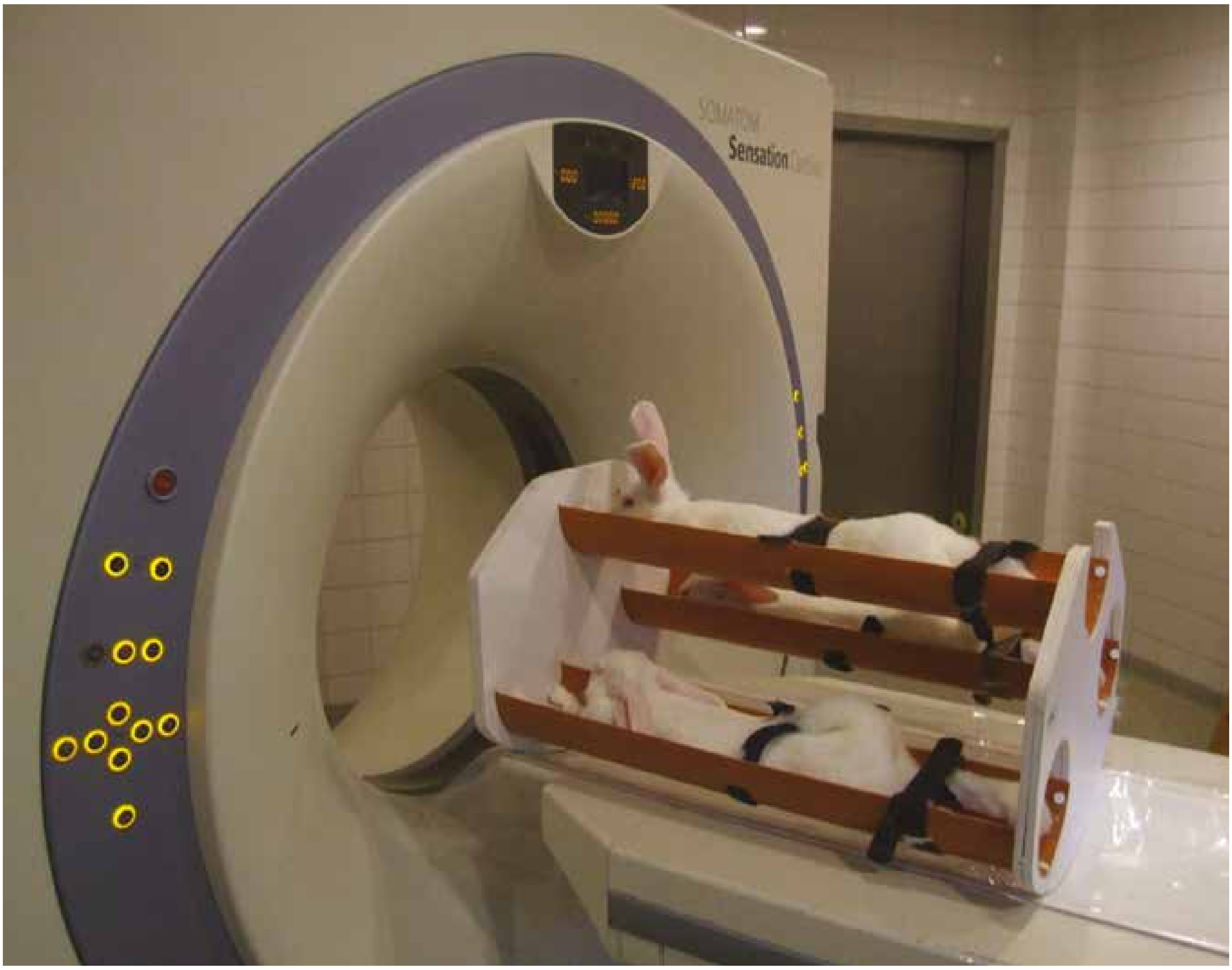


Figure 2. The thresholded images after axial morphology **(a)**; and the selection of the largest connected component **(b)**.

In practice, one Gabor-filter can enhance line segments of an angle. In order to enhance the contrast of line segments of any angle, *matched filtering* (Turin, 1960) is carried out. Basically, a matched filter is a set of filters (in our case Gabor-filters with different orientations). The application of the matched filter means the application of the filters in the set and the response of the matched filter is defined as the maximum response of the filters belonging to the set. In our case, the matched Gabor-filter is applied three times, in the axial, coronal and sagittal directions, respectively, and the filtered images are added to get the final filtered image. See Figure 1a and 1b for a CT image of a rabbit and the result of the filtering described above. Note that the body of the rabbit is touching the inner surface of the trough and the contrast of the trough and the body on the filtered image is better than that on the original image.

Figure 3. Multi-object CT scanning with rabbits.



The second step is *the thresholding of the filtered image* to identify the pixels of the trough. Since the intensity values of the voxels depend on the material of the trough and the parameters of the acquisition, one cannot use a fixed hard threshold. We have chosen Ridler's adaptive thresholding (Ridler and Calvard, 1978) and apply it slice-by-slice: let T_o denote a randomly selected initial threshold.

$$T_{n+1} = \frac{m_f(T_n) + m_b(T_n)}{2},$$

Then, the final threshold is selected by the iterative method where $m_f(T_n)$ and $m_b(T_n)$ denote the mean intensity of the foreground and background when T_n is used for thresholding.

See Figure 1b and 1c for the filtered and thresholded images. Note that there are several artifacts that thresholding cannot remove: the bones in the paws of the rabbit have similar intensities and are so close to the inner surface of the trough that they seem to be a connected object.

The third step of the proposed method involves mathematical morphology in the axial direction (Najman et. al., 2010). This is the step where we are heavily utilizing the concept that the cross section of the trough is supposed to vary slowly along the axial axis: the segments belonging to the animal are changing fast and some axial morphological opening followed by reconstruction can more-or-less completely remove them.

See Figure 2a for the results of morphological opening and reconstruction. It is easy to see that most of the artifacts are removed, only some elongated objects have remained in the middle of the image space.The fourth step is the extraction of the largest connected component as a binary mask covering the voxels of the trough (see Figure 2b).

The results obtained

The method was tested by the following protocol: three empty troughs (T1, T2, T3) were scanned (the axial sections are similar but slightly wounding and dilating); the mean volume of the slices and the corresponding standard deviations were determined; 5-5 images of the troughs containing rabbits were scanned, the troughs were extracted by the proposed method and the mean volumes of the slices and the corresponding standard deviations were determined. The parameters of the matched Gabor-filters applied to the axial slices are $\psi=0,\gamma=0.35,\lambda=12,\sigma=\lambda/3$, and the parameters of the filters applied to the coronal and sagittal slices differ only in $\gamma=0.175$. The size of the linear structuring element used for axial morphology is 27. The results are summarized in Table 1.

According to the results, the difference between the segmentation and the gold standard is approximately 5% and the standard deviations have the same orders of magnitude.

Table 1. The results of the quantitative evaluation of the proposed method.

Test case	**Mean T1 (mm^3)**	**Stdev T1 (mm^3)**	**Mean T2 (mm^3)**	**Stdev T2 (mm^3)**	**Mean T3 (mm^3)**	**Stdev T3 (mm^3)**
Gold standard	*2387.96*	*73.61*	*2403.82*	*66.58*	*2132.30*	*54.33*
#1	2296.14	44.47	2341.17	42.18	2066.70	72.10
#2	2303.87	43.25	2309.12	46.16	1996.58	62.73
#3	2228.63	40.61	2284.65	33.37	2049.05	79.94
#4	2191.97	65.28	2353.17	56.69	2086.20	81.25
#5	2291.18	76.10	2315.90	64.21	2220.99	55.77
Mean	*2262.36*	*53.94*	*2320.80*	*48.52*	*2083.90*	*70.36*
Error	*5.25%*		*3.45%*		*2.26%*	

The scientific conclusions

The proposed method is working, does not require any template or geometric model of the trough and the error rate is small enough to enable the applications of the method in practice.

Gabor-filters, thresholding methods and morphological operators are fast and powerful tools for the automated processing of 3D volumetric images.

The next steps

The filter response of Gabor-filters should be changed from inner product to Pearson-correlation coefficient to enable the absolute characterization of filter matching and the thresholding should be extended by some intelligent region growing algorithm. The testing protocol has to involve more troughs of various shapes, further test cases and parameters.

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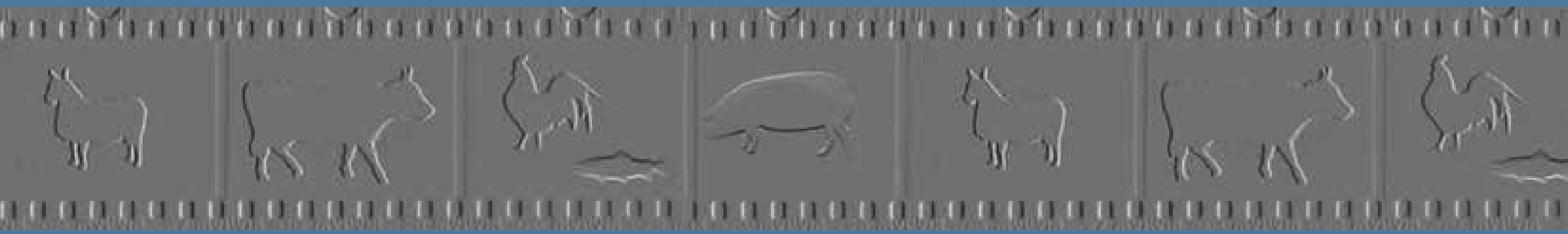
Workgroup 4

Kathy Peebles



Electronic Identification (EID) - From A Scottish Perspective

K. Peebles

Quality Meat Scotland, Rural Centre, Ingliston, Edinburgh, EH28 8NZ.

Value For Industry

EID can be used to:

- Provide traceability and proof of provenance.
- Measure animal performance at a farm level.
- Track individual animals through an abattoir.
- Deliver feedback to various sectors of the supply chain.

With a breeding flock of 2.8 million ewes there are tangible benefits to be had for the Scottish sheep industry.

Background

Sheep are an integral part of the Scottish Economy with 28% of all Scottish holdings involved in some form of sheep production and it is the largest sector of all the livestock enterprises. The sheep sector's contribution to the agricultural output of Scotland for the past year, as quoted by the Scottish Government, was £261 million and it continues to be the 2nd largest contributor of the red meat sectors. The stocking density varies throughout Scotland as is highly dependent on the topography. It ranges from >0 to 0.5 per hectare in the North and North West of the country to more than 3 animals per hectare in the flat arable areas of the southern areas.

Scotland, similar to other parts of the UK has previously had systems in place to trace batches of sheep and individual cattle. However, with the outbreak in 2001 of foot and mouth in the UK the paper to electronic based system called SAMU highlighted a time lag and the system was deemed to be unsatisfactory for the purposes of sheep traceability. (It is worth noting that since 2002 the Scottish auction markets have electronically reported the off farm, onto market movement for consigned animals and the off market and onward destination location movement, and the abattoirs have electronically reported the off farm and into abattoir movement on behalf of their supplying farmers for both cattle and sheep).

With the passing of Council Regulation EC 21/ 2004 by the European Commission to establish a system for the identification and registration of ovine and caprine animals, the decision was taken by the Scottish Government, after consultation with industry stakeholders, to implement a centralised electronic database, to track sheep movements in Scotland in real time. It is worth noting at this point that cattle are currently exempt from the requirement as there is already a paper based passport system with a centralised UK database. However, the Scottish view would be to centralise all sheep, pigs and cattle on a real time traceability database enabling faster tracking of animal movements in the event of a notifiable disease outbreak. The other aspect that needs to be clarified here for the reader is that the frequency of low frequency radio frequency identification (LF RFID) allowable in the UK is not the same as in other parts of the world i.e. New Zealand, as the frequency for the UK is set the lower rate of 134.2KHz.

Why Work is Needed

The regulation states that "the electronic identification for ovines and caprine animals has been developed to the stage where it can be applied". However, the EC funded IDEA trials from which this conclusion was drawn from were of a very limited nature. They did not take into account extensive hill flock production systems with semi-feral animals and a marketing system, in which there would be high throughput through the auctions markets and abattoirs, typical of the sheep industry in Scotland and other parts of the UK. Consequently Regulation EC 21/2004 does not provide a system capable of delivering 100% read rates for individual animals within a Scottish context.

Figure 1. A range of electronic tag readers.

The low frequency technology proscribed in 21/2004 is ideal for reading individual animals in a one by one system but is far less suitable for reading multiple animals simultaneously.

As a flock animal, sheep prefer to move as a group rather than individually. It is less stressful for the animal and is the least time consuming way of moving them, with one at a time reading in many situations being either impractical or impossible, particularly in high throughput marts and abattoirs. It is worth noting that the legislation was of particular concern as imposed additional costs on an industry where margins are, at the best of times, small and after the crash of 2001 were just starting to recover.

These costs were due to: expense of reading equipment; the additional labour time to record individual animals accurately. The threat of a loss of subsidy, which is a lifeline for many rural businesses and infrastructure, due to a failure to record individual animals accurately was problematic on such a large scale as human misreads are common. In Scotland, we have been fortunate that the Scottish Government has funded the Scoteid project to try and resolve some of the issues associated with EID implementation. It was also viewed from the start that this new type of technology could provide significant benefits as a farm management tool if the system could work.

The Issues

Since January 2010, under Scottish law, all sheep must be EID tagged. This decision was taken in the aftermath of research in 2008 into the development of a high throughput real-time recording system of individual sheep electronic identifications to a Central Database.

In 2009, the legislation was amended to allow a derogation for recording animal identifications which in Scotland are known as Critical Control Points (CCP) - these are the Auction Markets and Abattoirs. The decision was made after consultation with the industry not to exercise the derogation of the non-electronic ear tags, unlike other parts of the UK, as in Scottish terms this was viewed as not delivering a better system. However, Scotland in line with other parts of the UK did opt to use the what you see is what you get - WSIWYG format,that is to say that the number written on the outside of the electronic ear tag is the same as the number that the electronic reader will give without the need for a tag bucket.

With the formation of the CCP large volumes of sheep were able to be electronically read negating the need for individual farm businesses to purchase expensive reading equipment and allowing real time feedback. The volume of data this generated and continues to generate, also allows the Scoteid project to quantify the scale of the issues and to give up to date feedback to the industry.

These are:

- The quality of the electronic chip used
- The quality of the plastic used in the manufacturer of the ear tag
- The design of the ear tag
- Ear tag retention
- Quality control at the factory
- The practicalities of reading high volumes of sheep in a high throughput environment (and the legislation allows for no individual number on the outside of flock tag)
- Ear Tag lifespan
- Electronic reader and software compatibility
- The Financial Cost

However, as a farm management tool LF RFID has the potential to allow sheep keepers to better manage their flocks and to target where efficiencies in their flock management could be derived. It also has the potential to be able to deliver wider supply chain benefits as QMS in collaboration with Scoteid are working on systems to design a mechanism to feed data up and down the supply chain.

On Going Work

There are many reasons for the failure of tags to be read but the main one is still volume of read rate due to high throughput. Electronic read rates are dependent on the orientation of the ear tag to the actual reader i.e. individual reads tend to be higher than multiple reads as well as the factors listed below.

Quality Control. Other than the initial 50 "selected" ear tags sent to the competent authority there is no further statutory quality management requirement under EU 21/2004. The Scoteid project evaluates the different types of LF RFID sheep ear tags available to the farmer and have found the following reasons for lower read rates:

a) The electronic transponder has fallen out of the tag

b) The electronic transponder has been set at an incorrect frequency

c) The electronic transponder has been found coded with either the incorrect information re country, species etc. or there has been no data written on the transponder – it is blank

Internal Physical Damage. With some of the styles of ear tags licensed for use the transponder is sited in the pin of the ear tag and this can become damaged during insertion of the ear tag into the animal's ear and the farmer may not realise that this has happened. There may also be a failure to read as there may have been damage to the transponder during the manufacturing process which may only be detectable by using x-ray technology.

Retention of the ear tag in the animal's ear. Physical ear tags losses can occur through a number of factors and not just by the animal getting its ear caught in Rylock fencing or feed barriers. Button tags appear to be more prone to this type of problem than fold over tags. Improper application of the ear tag can be a major contributing factor along with time of year of application as tagging in the period when flies, and the famous Scottish midges (Cullicodes), are at their peak is not conducive to good tag retention but may be necessary as it coincides with peak movement periods to the livestock sales. There may also be problems where the fold of the ear tag has broken

Manufacturing design. Over the past 5 years the Scoteid project has fed information back to the tag manufacturers to show the types of problems that are occurring and have produced a league table of offenders. Poor design particularly of the pins holding the two parts of the ear tag together appear to be amongst the main issues with problems associated with the type of plastic used in the tag manufacturing process which has then become brittle and breaks apart when exposed to weather related problems such as sunlight, frosty, snowy and wet conditions which are the norm in a Scottish sheep calendar year.

Figure 2. Electronic tag reading.

The next steps

In the past you had to visually try to read small print on an ear tag with the resulting mis-matches that occurred and until the implementation of the LFRFID there was not an easy way of measuring performance from large numbers of sheep in a commercial situation. Feedback from the abattoir to the farm is currently in the form of a batch reporting system. This does not allow producers to check an individual lamb's performance with the weight and grade that the lamb has attained. QMS, in-conjunction with Scoteid and one of our larger lamb processors, are working on a system that could be rolled out throughout Scotland in meat plants that EUROP grid grade lambs. This system could enable consignors to see exactly what their individual lambs have achieved and could also enable the store producer (the one who breeds the lamb and then sells it to a finisher as they do not have land suitable for finishing) to see how their animals have performed within the supply chain.

This system could also deliver wider benefits to the meat plants as they would know exact farms of origin of the Scottish animals supplied. The Scoteid Hub can already give identify the locations of every animal during its lifetime at the click of a few buttons on a computer but until individual feedback from the abattoir is a reality there is a black hole in the information flow that could help farmers with their breeding decisions.

Liver fluke costs. Condemned livers that could be utilised by the human food chain are a waste and disposal costs. So having a better mechanism to identify where problems like this are arising and having a better feedback mechanism to both the consignor and the breeder is a real benefit to the industry and could only be made possible through the Scoteid Data Hub.

One of the other drawbacks with the current legislation is that there is currently no mechanism in place to update the technology. Even before the Regulation implementing LF RFID was in place there were other technologies being more widely used by the food distribution sectors that were more robust, more effective and cheaper to purchase namely UHF. As mentioned earlier there is no legislation currently in place for the electronic identification of cattle. However, it is being considered by the European Commission. Scoteid has been researching the pros and cons of this type of technology and has developed, in-conjunction with some of the tag manufacturers, UHF cattle tags that are readable at a distance of up to 6 metres depending on the frequency of setting. This type of ear can also allow farmers to be able to write management information to the tag. Imagine a system where no more soggy papers at the cattle crush or a tedious means of trying to identify previous history for that animal without pieces of paper...... This takes the cattle and, if legislation would allow, the sheep sectors to a whole new world of up to date management information flow.

Acknowledgements

This work was primarily funded by Scottish Government through the Scoteid project with input from QMS and other parts of the supply chain.

The industry is very grateful to the research undertaken by the Scoteid development team led by Hamish Stuart of Black Isle Technical Services.

References

Scoteid **www.scoteid.com**
Quality Meat Scotland **www.qmscotland.co.uk**

Poster Competition: The winner

Torunn Aasmundstad, Norsvin

norsvin

Can computed tomography of live animals be a tool for scoring osteochondrosis in a pig breeding program?

Torunn Aasmundstad[1,2], Jørgen Kongsro[1] and Odd Vangen[2]
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Value for industry:
- *In vivo* assessment of osteochondrosis by use of computed tomography (CT) can enable phenotyping of live animals.
- A non-invasive method will improve genetic gain of the trait by increasing the accuracy of the breeding values.

Background:
Osteochondrosis (OC) is a disturbance in the process of enchondral ossification affecting individuals at early age (before regression of blood supply from the growth cartilage).The condition can lead to lameness, and is traditionally assessed *post mortem* and in two ways; 1) macroscopically inspection of the joint cartilage surface or 2) by sectioning the joint in slabs and scoring the slabs. If CT could be used as a tool for assessing OC *in vivo*, this would facilitate phenotypes on the breeding animal itself and hence enable better control of OC in a breeding population.

Objective:
- Establish a method for assessing OC by means of CT (OCwCT) on eight anatomical locations (medial and lateral condyles at the distal end of the humerus or the femur of the right and left leg).
- Calculate heritabilities for the traits.
- Estimate the genetic correlations among the traits.

The method used:
In the pig breeding program of Norsvin, all boars are subjected to CT scan at end of boar test (~100 kg). The routine was implemented in 2008, and since then >17 000 boars has been scanned. From this material, 2273 boars were assessed for OCwCT on eight anatomical locations and the sum score for OC for each animal (OCT). The pixel size of the images was 0.93 mm^2 and the slice thickness was 1.25 mm. The high resolution permits 3D visualisation as can be seen in fig 1 and fig 2. The material was assessed by one assessor and took about 3-4 minutes for each animal. Signs of OC interpreted as changes in the interface between the joint cartilage and the encondral bone plate were seen as areas with increased radiolucency. For viewing purposes the software OsiriX was used. A threshold window level of 400 HU and a window width of 400 HU were applied to enable best contrast/detail level for the tissue of interest. A grading scale was developed and is shown in fig 3. For the preliminary statistical handling of the phenotypes the SAS/STAT® software were used while estimates of genetic variance components was output from the DMU 6.7/AI-REML software package.

Acknowledgements:
The study presented is a part of the «Robust pig» project funded by the Research Council of Norway (project no: 199598).

Results

Figure 1: 3D model based on CT of posterior end of a boar. The boar shows signs of OC on femur medial condyle.

Figure 2: 3D model based on CT of the left femur of a boar. OC lesions can be seen both on the medial and lateral condyle.

Figure 3: Scoring of osteochondrosis in the distal humerus and femur in pigs with computed tomography. 0 denotes no lesion, 1 denotes minimal lesion, 2 denotes moderate lesion, 3 denotes considerable lesion and 4 denotes small dissecans. 5 denotes large dissecans, not seen in the material. All images are from the right medial condyle of humerus.

Figure 4: Prevalence of OCwCT for each anatomical location. Anatomical abbrevations: First letter position H=Humerus, F=Femur. Second letter position: R=right, L=left. Third letter position: M=medial, L=lateral.

Figure 5: Distribution (percent) of boars within each class of OCT score.

Table 1: Heritabilities and genetic correlations for the individual locations and the sum of the locations. All estimates are from bivariate analysis. N.E. indicates analysis that did not converge within the set criterion.

	OCT	HRM	HLM	FRM	FRL	FLM	FLL
OCT	**0.29(0.06)**	0.92(0.04)	0.96(0.03)	0.65(0.13)	0.62(0.17)	0.63(0.14)	0.56(0.41)
HRM		**0.28(0.06)**	0.99(0.03)	0.34(0.19)	0.37(0.24)	0.17(0.22)	0.06(0.45)
HLM			**0.27(0.06)**	0.33(0.2)	0.34(0.24)	0.35(0.21)	0.39(0.47)
FRM				**0.12(0.04)**	0.22(0.31)	0.89(0.13)	-0.15(0.6)
FRL					**0.07(0.03)**	0.04(0.37)	N.E
FLM						**0.09(0.04)**	0.23(0.57)
FLL							**0.02(0.03)**

The scientific conclusions:
- CT may be used as a tool for assessing osteochondrosis *in vivo*.
- The trait «sum score of osteochondrosis» (OCT) has a medium heritability (see table 1).
- The genetic correlations between the eight individual locations investigated and OCT are high, suggesting that a selection for reduced OCT would reduce incidence at all locations.
- As the selection candidates are subjected to CT, phenotype from the candidates will improve accuracy of the breeding values and hence increase potential of genetic gain.

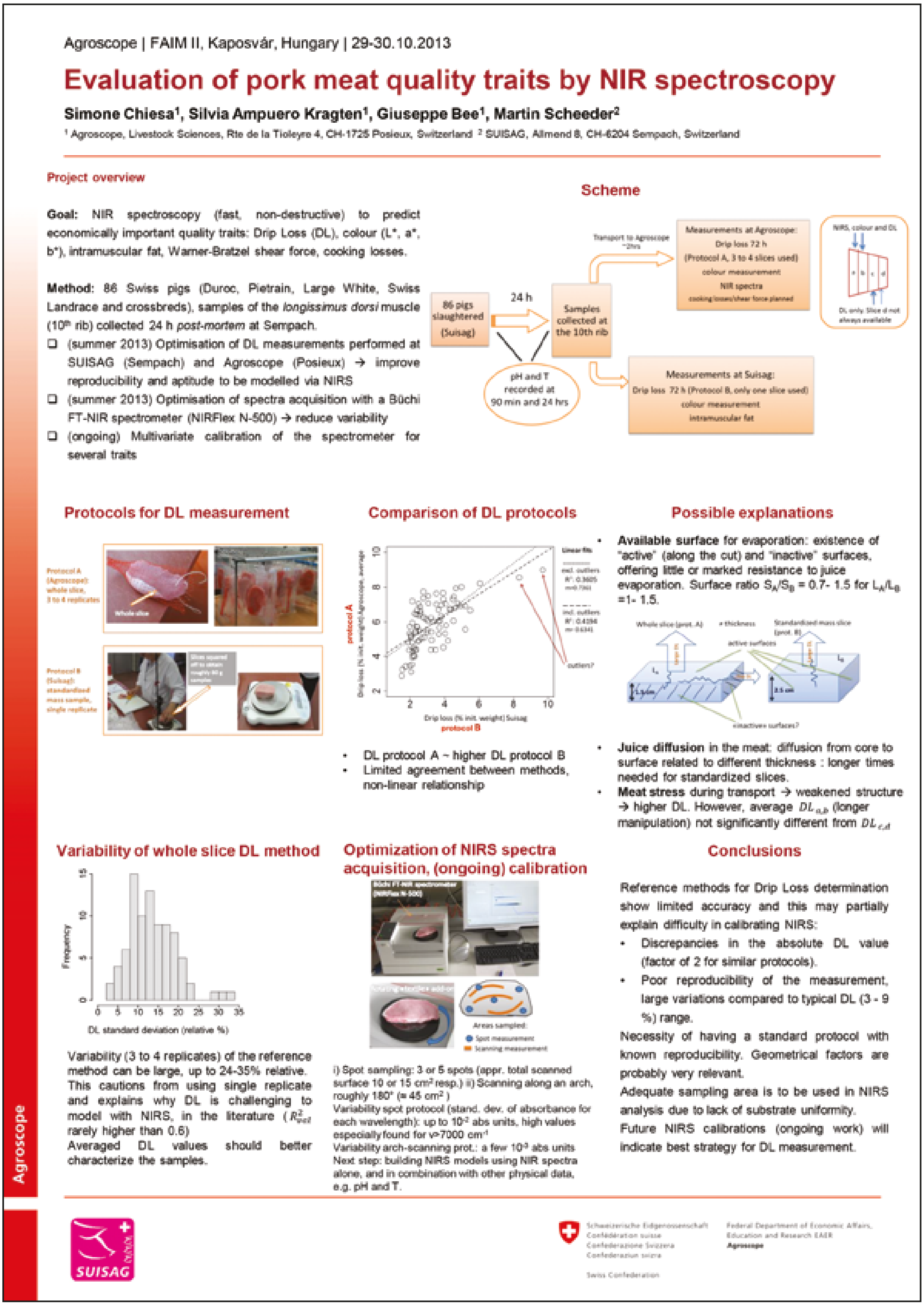
The next steps:
- Investigate possibility for semi-automatic phenotyping of OC.
- Develop a technique to distinguish the lesions subjected to repair from that of a loosening bone fragment.
- Implement OC as a selection criterion in a pig breeding goal.
- Use CT to study longitudinal development of OC lesions in growing pigs.
- Investigate phenotypic and genetic relationship between conformation traits and OC.
- Investigate potential genetic link between OC and sow removal.

PhD-student
Torunn Aasmundstad

Layout og trykk: www.gshamar.no

Poster Competition: 2nd place

Simone Chiesa, Agroscope



Agroscope | FAIM II, Kaposvár, Hungary | 29-30.10.2013

Evaluation of pork meat quality traits by NIR spectroscopy

Simone Chiesa[1], Silvia Ampuero Kragten[1], Giuseppe Bee[1], Martin Scheeder[2]

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Project overview

Goal: NIR spectroscopy (fast, non-destructive) to predict economically important quality traits: Drip Loss (DL), colour (L*, a*, b*), intramuscular fat, Warner-Bratzel shear force, cooking losses.

Method: 86 Swiss pigs (Duroc, Pietrain, Large White, Swiss Landrace and crossbreds), samples of the *longissimus dorsi* muscle (10th rib) collected 24 h *post-mortem* at Sempach.

- ☐ (summer 2013) Optimisation of DL measurements performed at SUISAG (Sempach) and Agroscope (Posieux) → improve reproducibility and aptitude to be modelled via NIRS
- ☐ (summer 2013) Optimisation of spectra acquisition with a Büchi FT-NIR spectrometer (NIRFlex N-500) → reduce variability
- ☐ (ongoing) Multivariate calibration of the spectrometer for several traits

Scheme

86 pigs slaughtered (Suisag)

24 h

Samples collected at the 10th rib

pH and T recorded at 90 min and 24 hrs

Transport to Agroscope ~2hrs

Measurements at Agroscope:
Drip loss 72 h
(Protocol A, 3 to 4 slices used)
colour measurement
NIR spectra
cooking losses/shear force planned

Measurements at Suisag:
Drip loss 72 h (Protocol B, only one slice used)
colour measurement
intramuscular fat

Protocols for DL measurement

Protocol A (Agroscope): whole slice, 3 to 4 replicates

Protocol B (Suisag): standardized mass sample, single replicate

Comparison of DL protocols

- DL protocol A ~ higher DL protocol B
- Limited agreement between methods, non-linear relationship

Possible explanations

- **Available surface** for evaporation: existence of "active" (along the cut) and "inactive" surfaces, offering little or marked resistance to juice evaporation. Surface ratio S_A/S_B = 0.7- 1.5 for L_A/L_B =1- 1.5.

- **Juice diffusion** in the meat: diffusion from core to surface related to different thickness : longer times needed for standardized slices.
- **Meat stress** during transport → weakened structure → higher DL. However, average $DL_{a,b}$ (longer manipulation) not significantly different from $DL_{c,d}$

Variability of whole slice DL method

Variability (3 to 4 replicates) of the reference method can be large, up to 24-35% relative. This cautions from using single replicate and explains why DL is challenging to model with NIRS, in the literature (R^2_{val} rarely higher than 0.6)
Averaged DL values should better characterize the samples.

Optimization of NIRS spectra acquisition, (ongoing) calibration

i) Spot sampling: 3 or 5 spots (appr. total scanned surface 10 or 15 cm² resp.) ii) Scanning along an arch, roughly 180° (≈ 45 cm²)
Variability spot protocol (stand. dev. of absorbance for each wavelength): up to 10^{-2} abs units, high values especially found for ν>7000 cm⁻¹
Variability arch-scanning prot.: a few 10^{-3} abs units
Next step: building NIRS models using NIR spectra alone, and in combination with other physical data, e.g. pH and T.

Conclusions

Reference methods for Drip Loss determination show limited accuracy and this may partially explain difficulty in calibrating NIRS:

- Discrepancies in the absolute DL value (factor of 2 for similar protocols).
- Poor reproducibility of the measurement, large variations compared to typical DL (3 - 9 %) range.

Necessity of having a standard protocol with known reproducibility. Geometrical factors are probably very relevant.

Adequate sampling area is to be used in NIRS analysis due to lack of substrate uniformity.

Future NIRS calibrations (ongoing work) will indicate best strategy for DL measurement.

Agroscope

SUISAG

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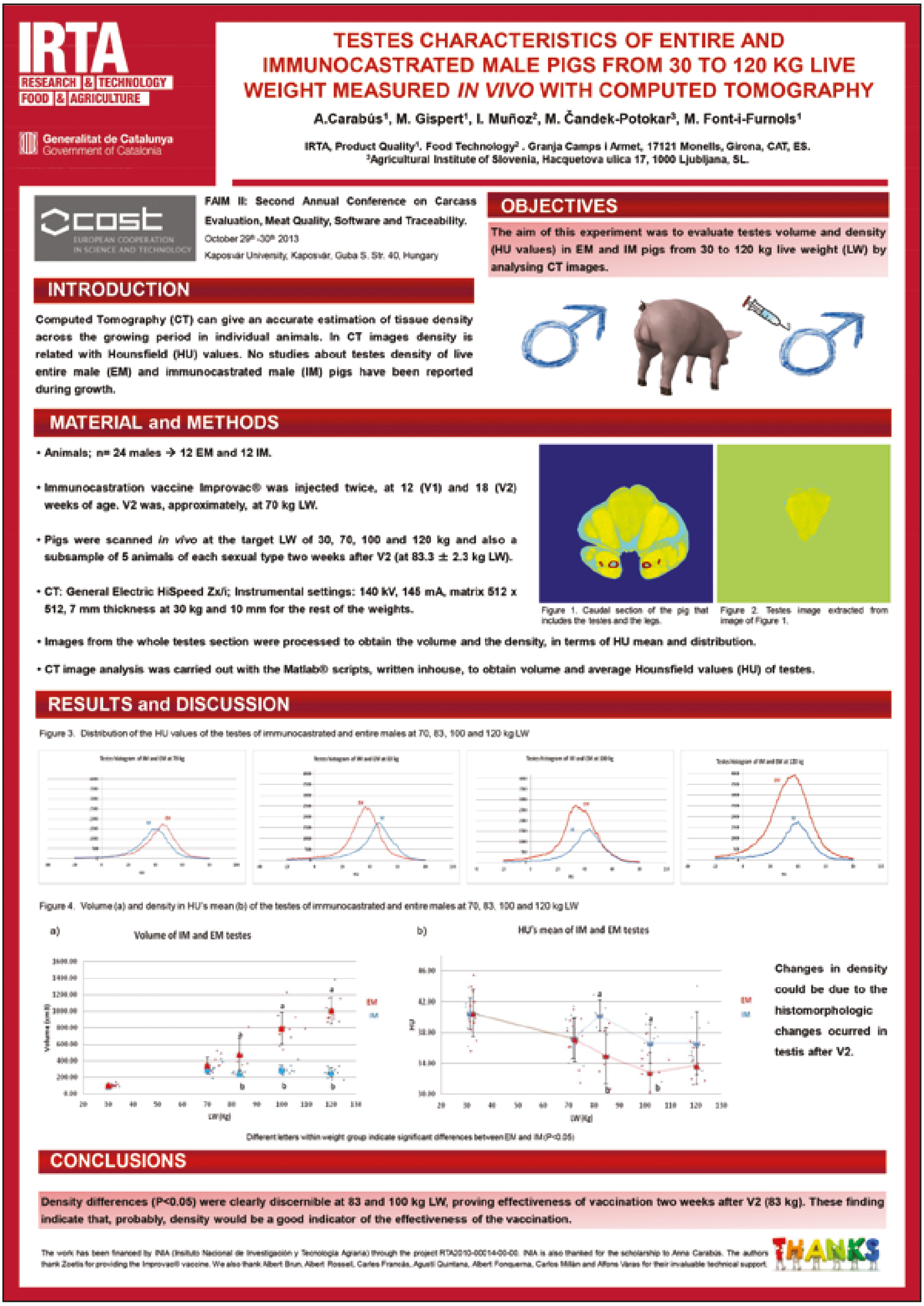
Confederaziun svizra

Swiss Confederation

Federal Department of Economic Affairs, Education and Research EAER
Agroscope

Poster Competition: 3rd place

Anna Carabús, IRTA



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RESEARCH & TECHNOLOGY
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Generalitat de Catalunya
Government of Catalonia

TESTES CHARACTERISTICS OF ENTIRE AND IMMUNOCASTRATED MALE PIGS FROM 30 TO 120 KG LIVE WEIGHT MEASURED *IN VIVO* WITH COMPUTED TOMOGRAPHY

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cost
EUROPEAN COOPERATION IN SCIENCE AND TECHNOLOGY

FAIM II: Second Annual Conference on Carcass Evaluation, Meat Quality, Software and Traceability.
October 29th -30th 2013
Kaposvár University, Kaposvár, Guba S. Str. 40, Hungary

OBJECTIVES

The aim of this experiment was to evaluate testes volume and density (HU values) in EM and IM pigs from 30 to 120 kg live weight (LW) by analysing CT images.

INTRODUCTION

Computed Tomography (CT) can give an accurate estimation of tissue density across the growing period in individual animals. In CT images density is related with Hounsfield (HU) values. No studies about testes density of live entire male (EM) and immunocastrated male (IM) pigs have been reported during growth.

MATERIAL and METHODS

- Animals; n= 24 males → 12 EM and 12 IM.
- Immunocastration vaccine Improvac® was injected twice, at 12 (V1) and 18 (V2) weeks of age. V2 was, approximately, at 70 kg LW.
- Pigs were scanned *in vivo* at the target LW of 30, 70, 100 and 120 kg and also a subsample of 5 animals of each sexual type two weeks after V2 (at 83.3 ± 2.3 kg LW).
- CT: General Electric HiSpeed Zx/i; Instrumental settings: 140 kV, 145 mA, matrix 512 x 512, 7 mm thickness at 30 kg and 10 mm for the rest of the weights.

Figure 1. Caudal section of the pig that includes the testes and the legs.

Figure 2. Testes image extracted from image of Figure 1.

- Images from the whole testes section were processed to obtain the volume and the density, in terms of HU mean and distribution.
- CT image analysis was carried out with the Matlab® scripts, written inhouse, to obtain volume and average Hounsfield values (HU) of testes.

RESULTS and DISCUSSION

Figure 3. Distribution of the HU values of the testes of immunocastrated and entire males at 70, 83, 100 and 120 kg LW

Figure 4. Volume (a) and density in HU's mean (b) of the testes of immunocastrated and entire males at 70, 83, 100 and 120 kg LW

Changes in density could be due to the histomorphologic changes ocurred in testis after V2.

Different letters within weight group indicate significant differences between EM and IM (P<0.05)

CONCLUSIONS

Density differences (P<0.05) were clearly discernible at 83 and 100 kg LW, proving effectiveness of vaccination two weeks after V2 (83 kg). These finding indicate that, probably, density would be a good indicator of the effectiveness of the vaccination.

The work has been financed by INIA (Instituto Nacional de Investigación y Tecnología Agraria) through the project RTA2010-00014-00-00. INIA is also thanked for the scholarship to Anna Carabús. The authors thank Zoetis for providing the Improvac® vaccine. We also thank Albert Brun, Albert Rossell, Carles Francàs, Agustí Quintana, Albert Fonquerna, Carlos Millán and Alfons Varas for their invaluable technical support.

THANKS

Poster Competition: 4th place

Philippa Morrison, University of Liverpool

Relationships between body condition score and regional adipose depots in the horse

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Introduction & Hypothesis

- The EU has around **6 million** feral, leisure, performance and food production horses.
- **Equine body composition** is critically important to all sectors but methods to evaluate body composition in living animals are either subjective and imprecise (e.g. body condition scoring, BCS) (Dugdale et al., 2012) or objective but limited to research-specific applications (e.g Deuterium oxide) (Dugdale et al., 2011).
- **Whole animal imaging capabilities for *Equidae* are not available.**
- **Over 50%** of UK horses and ponies are **overweight or obese** (Harker et al., 2011), which impacts negatively on carcass composition, performance and welfare.
- Internal adipose depots have negative associations with health but cannot currently be evaluated (Carey et al., 1996).

HYPOTHESIS: Developing *post-mortem* scoring systems to evaluate regional adipose tissue depots would improve our understanding of any associations between regional fat deposition and *ante-mortem* evaluations of BCS and animal phenotype.

Fig 1. Medial and lateral view equine carcass

Methods

Mesenteric fat		
Grade	**Description**	**Image**
1	No/minimal fat visible.	
2	Fat in the immediate vicinity of the superior mesenteric vessels (SMV's) but arterial arcades still clearly visible.	
3	Distinct fat deposits around and beginning to fill the spaces between SMV's. SMV's partially obscured by fat.	
4	Extensive accumulations of fat largely obscuring and filling spaces between most arcades of the SMV's.	
5	Mesenteric peritoneum and SMV's completely obscured by fat.	

1. Adipose Depot-specific Scoring Systems: Anatomically defined photographic images of omental, mesenteric, epicardial and rump adipose depots from 40 animals across the range of BCS, were photographed and ranked to develop **depot-specific, 5 point scoring systems** (0 = no fat visible; 5 = maximal fat present) (e.g. Fig. 2). Depths (+/-1mm) of the nuchal crest and abdominal retroperitoneal adipose depots were recorded at depot midpoints on the split carcasses. The range of recorded depths were uniformly stratified and strata were used as the basis of 5 point scores for crest and retroperitoneal adipose depots (1 = least; 5 = greatest).

2. Data Collection

- 114 animals (75 mares, 37 geldings, 2 stallions), euthanased for non-research purposes and representative of the UK abattoir population on the basis of carcass weight and gender, were evaluated *ante-mortem* and again, immediately *post-mortem*.
- ***Ante-mortem***: Age in years (passport & teeth), estimated withers height, breed-type, gender, and BCS (1-9; where 1 = extremely thin and 9 = obese, Kohnke, 1992).
- ***Post-mortem***: Omental, mesenteric, epicardial, rump, nuchal crest and abdominal retroperitoneal scores (above) and carcass weight.
- Data analyses used STATA 12 (StataCorp, U.S.A) Three, multi-variable random effects linear regression models with BCS as the outcome variable were fitted with breed considered as a random effect. The range of explanatory variables differed for each model. **Model 1;** included all phenotypic data and fat scores. **Model 2;** only included external and internal fat scores. **Model 3;** only included the phenotypic measures.

Fig 2. Mesenteric adipose tissue scores with associated descriptors and typical reference images. Comparable scoring systems were also developed for the omental, epicardial and rump fat depots.

Results

Table 1. Mixed effects multivariable logistic regression model including variables associated with BCS. Breed is considered as a random effect. CI, confidence interval.

	MODEL 1 (Adj. R^2 = 0.68) *(Breed variance = 0.99)* *(95% CI = 0.18 to 0.55)*			**MODEL 2 (Adj. R^2 = 0.57)** *(Breed variance = 0.26)* *(95% CI = 0.07 to 1.00)*			**MODEL 3 (Adj. R^2 = 0.42)** *(Breed variance = 0.15)* *(95% CI = 0.04 to 0.57)*		
Variable	Estimate β	95% CI	*P* value	Estimate β	95% CI	*P* value	Estimate β	95% CI	*P* value
Height (in cm/10)	-0.47	-0.69 to -0.25	< 0.001				-0.75	-1.01 to -0.50	< 0.001
Age (years)	-0.02	-0.05 to 0.004	0.01				-0.02	-0.05 to 0.001	0.26
Gender (male or female)	0.27	-0.06 to 0.59	0.11				0.28	-0.12 to 0.69	0.17
Carcass weight (kg/10)	0.06	0.02 to 0.10	0.005				0.14	0.10 to 0.18	< 0.001
Mesenteric fat score	-0.13	-0.33 to 0.06	0.19	-0.15	-0.36 to 0.006	0.16			
Epicardial fat score	0.05	-0.12 to 0.22	0.55	0.05	-0.13 to 0.23	0.58			
Rump fat score	0.14	-0.05 to 0.32	0.15	0.24	0.05 to 0.43	0.001			
Omental fat score	0.15	-0.05 to 0.35	0.14	0.21	0.002 to 0.42	0.05			
Crest fat depth score	0.35	0.16 to 0.55	< 0.001	0.44	0.23 to 0.64	< 0.001			
Retroperitoneal fat depth score	0.50	0.28 to 0.72	< 0.001	0.44	0.21 to 0.66	< 0.001			
Baseline	8.17	4.55 to 11.79	< 0.001	2.05	1.15 to 2.96	< 0.001	14.94	12.09 to 17.79	< 0.001

- Means were: BCS, 5.02 (range 2.25 - 8.25); Age,10.82 (range 1.50 – 33.00 years) withers height, 152.8cm (range 102.0 – 178.0cm).
- Models 1 & 2 (Table 1) confirmed strong positive associations between crest and retroperitoneal fat scores and BCS, both in magnitude and statistical significance.
- Scores for both rump and omental fats were positively associated with BCS in Models 1 & 2, although the magnitude of the associations were less than noted for crest and retroperitoneal depots. Conversely, mesenteric fat scores were negatively associated with and BCS, while epicardial fat scores were independent of BCS in both Models.
- Inspection of Models 1 & 2 demonstrates the strong confounding effect of visible external phenotypic data on the associations between internal and external fats with BCS.
- Model 3 indicated previously unreported impacts of phenotype, specifically animal size, age and gender on BCS predictions.
- Appraisal of predicted changes in BCS for each regional fat score (Fig.3), clearly indicated that for the majority of depots (omental, rump, retroperitoneal and crest), there was a trend for BCS to increase with unit increases in specific fat scores. Conversely, on the basis of these data, BCS had a tendency to decrease as mesenteric fat scores increased.

Fig 3. Marginal means (95% CI) illustrating predicted changes in BCS with fat scores (retroperitoneal & crest **(A)**, epicardial & rump **(B)**, omental & mesenteric **(C)** fats).

Conclusion

- This study demonstrates the first manual scoring method for fat analysis in the horse.
- Clear differences in trelationships between BCS and the 6 regional fat deposits evaluated suggest different functional roles for in the individual adipose tissues.
- Nuchal crest and ventro-abdominal retroperitoneal fat scores were strongly associated with BCS which were not confounded by animal phenotype.
- The strong confounding influence of phenotype on rump fat scores is important because rump scores have been used as the basis of calculations of total body fat across breeds.
- There is now an urgent need for image-based methods for fat scoring in the horse.

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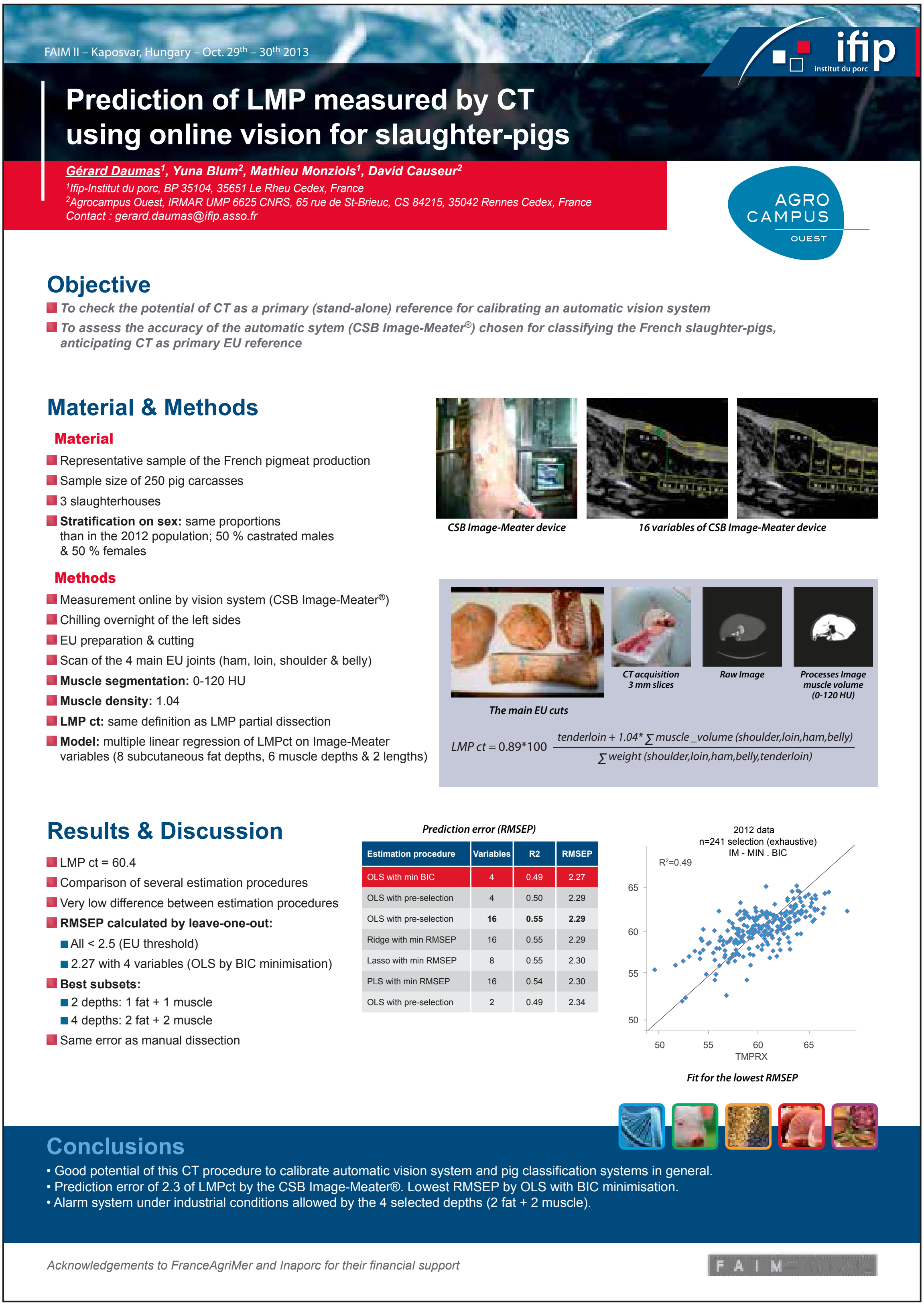
Poster Competition: 5th place =

Gerard Daumas, IFIP-Institut du Porc



FAIM II – Kaposvar, Hungary – Oct. 29th – 30th 2013

Prediction of LMP measured by CT using online vision for slaughter-pigs

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Objective

- *To check the potential of CT as a primary (stand-alone) reference for calibrating an automatic vision system*
- *To assess the accuracy of the automatic sytem (CSB Image-Meater®) chosen for classifying the French slaughter-pigs, anticipating CT as primary EU reference*

Material & Methods

Material

- Representative sample of the French pigmeat production
- Sample size of 250 pig carcasses
- 3 slaughterhouses
- **Stratification on sex:** same proportions than in the 2012 population; 50 % castrated males & 50 % females

CSB Image-Meater device

16 variables of CSB Image-Meater device

Methods

- Measurement online by vision system (CSB Image-Meater®)
- Chilling overnight of the left sides
- EU preparation & cutting
- Scan of the 4 main EU joints (ham, loin, shoulder & belly)
- **Muscle segmentation:** 0-120 HU
- **Muscle density:** 1.04
- **LMP ct:** same definition as LMP partial dissection
- **Model:** multiple linear regression of LMPct on Image-Meater variables (8 subcutaneous fat depths, 6 muscle depths & 2 lengths)

The main EU cuts

CT acquisition
3 mm slices

Raw Image

Processes Image
muscle volume
(0-120 HU)

$$LMP\ ct = 0.89*100\ \frac{tenderloin + 1.04*\sum muscle\_volume\ (shoulder,loin,ham,belly)}{\sum weight\ (shoulder,loin,ham,belly,tenderloin)}$$

Results & Discussion

- LMP ct = 60.4
- Comparison of several estimation procedures
- Very low difference between estimation procedures
- **RMSEP calculated by leave-one-out:**
  - All < 2.5 (EU threshold)
  - 2.27 with 4 variables (OLS by BIC minimisation)
- **Best subsets:**
  - 2 depths: 1 fat + 1 muscle
  - 4 depths: 2 fat + 2 muscle
- Same error as manual dissection

Prediction error (RMSEP)

Estimation procedure	Variables	R2	RMSEP
OLS with min BIC	4	0.49	2.27
OLS with pre-selection	4	0.50	2.29
OLS with pre-selection	**16**	**0.55**	**2.29**
Ridge with min RMSEP	16	0.55	2.29
Lasso with min RMSEP	8	0.55	2.30
PLS with min RMSEP	16	0.54	2.30
OLS with pre-selection	2	0.49	2.34

Fit for the lowest RMSEP

Conclusions

- Good potential of this CT procedure to calibrate automatic vision system and pig classification systems in general.
- Prediction error of 2.3 of LMPct by the CSB Image-Meater®. Lowest RMSEP by OLS with BIC minimisation.
- Alarm system under industrial conditions allowed by the 4 selected depths (2 fat + 2 muscle).

Acknowledgements to FranceAgriMer and Inaporc for their financial support

Poster Competition: 5th place =

A. Vautier, IFIP-Institut du Porc

FAIM II second annual conference: KAPOSVÁR, oct. 29th - 30th 2013, Hungary

Near infrared spectroscopy (NIRS) as a tool for predicting the technological quality of ham

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APRIVIS

Introduction

Muscle quality is a key factor in the French cooked ham industry. Phosphates and carraghenans are not allowed in the Jambon Cuit Supérieur process, and the ultimate pH is routinely used to control the quality of hams. Due to robustness issues, the processed meat industry is looking for an alternative to pH. The aim of this study was to test the feasability of the use of NIRS as a routine technique to predict cooking yield of hams. The precision of NIRS calibration to predict PSE-Like zone classification was evaluated too.

Material & Methods

- 110 deboned hams individually processed following industrial standards:

- ASDI spectrocolorimeter Labspec5000 (350-2500nm)
- Contact and insertion probes:

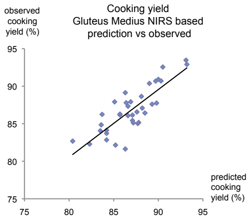
Estimation procedure	**Surface** 	**insertion** 
Muscle	*Semimembranosus* *Gluteus Medius* *Vastus Medialis* *Biceps Femoris* *Semitendinosus*	*Semimembranosus*

- Variable of interest: cooking yield (m=87.6, sd=3.6) and PSE-like zones
- PLS calibration modelling with cross-validation (n=74) followed by external validation (n=36)

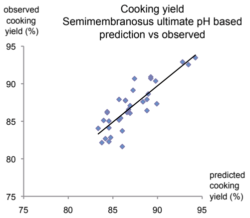
Results

Best external validation results for cooking yield were obtained with NIRS calibration on the *Gluteus Medius* cross section, which presents an excellent bone-in and deboned availability. Predicted/observed cooking yield correlation and predicting error (r=0.82, error=1.78) show a similar precision of NIRS compared to the *Semimembranosus* ultimate pH based prediction (r=0.85, error=1.66).

- *Gluteus Medius* external validation results for the prediction of the cooking yield with NIRS vs *Semimembranosus* ultimate pH based prediction (*second order polynomial model*):



r = 0.82 error = 1.78



r = 0.85 error = 1.66

- Other Muscles external validation results for the prediction of the **cooking yield:**

Probe	**Muscle**	**NIRS predicted / observed cooking yield (r =)**	**Error**
Surface	*Vastus Lateralis*	**0.60**	**2.52**
	Biceps Femoris	**0.53**	**2.64**
	Semitendinosus (*internal surface*)	**0.55**	**2.61**
	Semitendinosus (*external surface*)	**0.28**	**3.00**

NIRS classification of hams according to the PSE-like zone defect (presence / abscence) showed again a good level of precision for external validation with 84% of correct classification on deboned hams (surface probe) and 77% of correct classification on bone-in hams (insertion probe).

- External validation results for the prediction of the **PSE-like zones** classification (presence / abscence) on raw meat:

Probe	**Muscle**	**Correctly classified (%)**	**% false positive / % false negative**
Surface	*Gluteus Medius*	**60**	**77 / 23**
	Semimembranosus (*internal surface*)	**84**	**57 / 43**
Insertion	*Semimembranosus* (*muscle inside*)	**77**	**65 / 35**

Conclusions

External validation test confirms that visible and near infrared spectroscopy is an efficient alternative to pHu for cooking yield prediction. Developpements are still needed for a PSE-Like zone classification based on NIRS and using an insertion probe to recover the precision level of the surface probe.

FAIM

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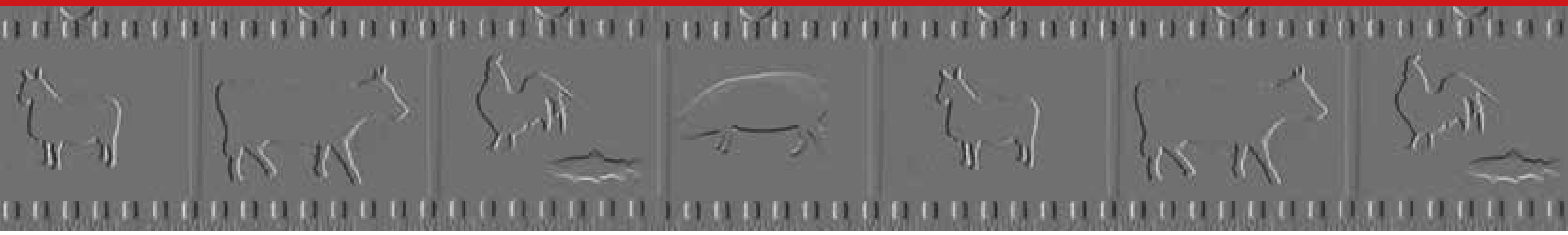

About this book

This book comprises reports of some of the activities of the COST action FAIM during 2012-2013. The papers contained in the book were presented at the FAIM meeting held in Kaposvár, Hungary in October 2013.

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FA2571_11/13

ISBN 978-0-9570709-9-8