

FAIM

Farm Animal IMaging

FARM ANIMAL IMAGING

DUBLIN 2012



C. Maltin, C. Craigie and L. Bünger

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Foreword

COST ACTION FA1102: FAIM (FARM ANIMAL IMAGING)

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What is COST?

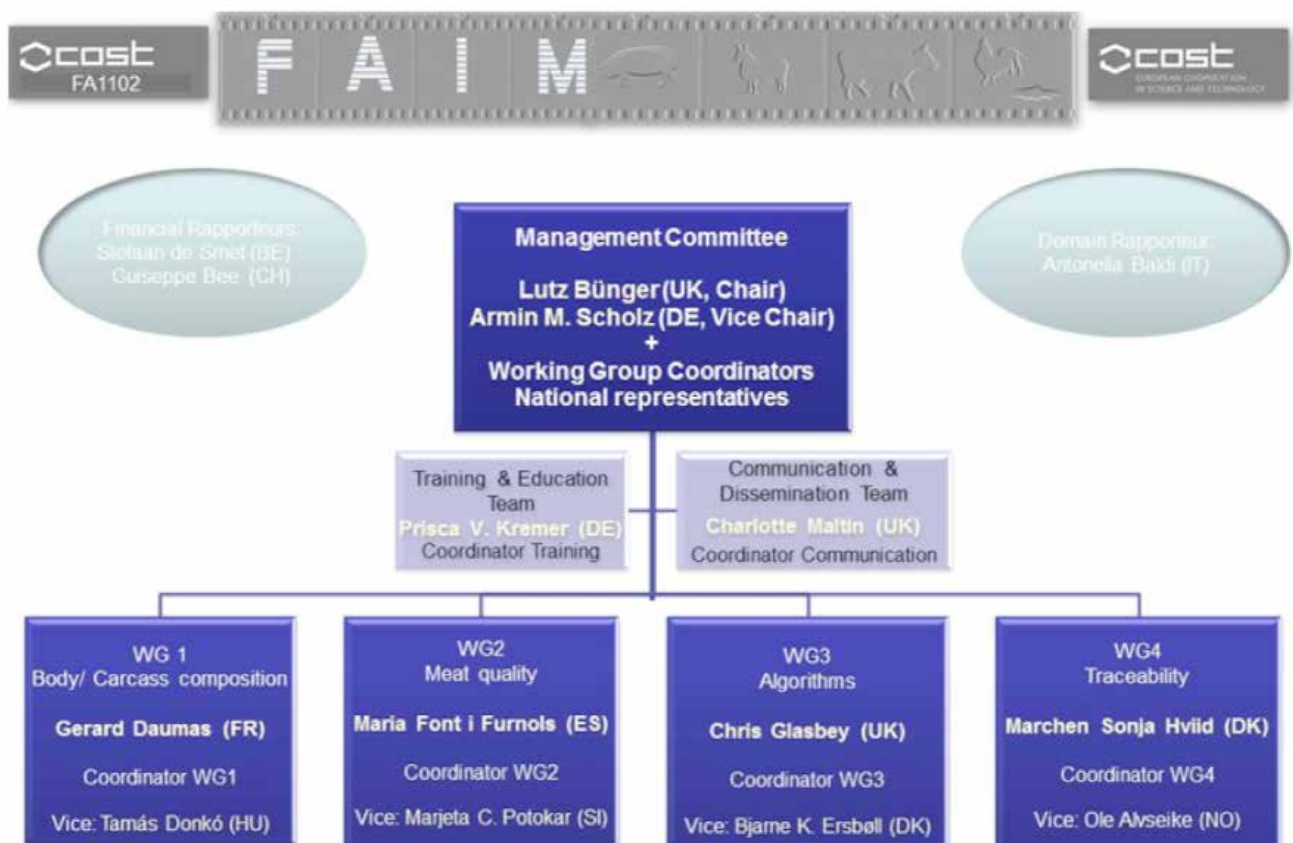
COST is a flexible, fast and efficient intergovernmental framework for European Cooperation in Science and Technology, allowing the coordination of nationally-funded research on a European level with a very specific mission and goal. It allows bringing good scientists and representatives of the industry together under light strategic guidance. COST is based on networks, called COST Actions, centred around research projects in fields that are of interest to at least five COST countries. Thereby COST contributes to reducing the fragmentation in European research investments and opening the European Research Area to cooperation worldwide. COST acts as a precursor of advanced multidisciplinary research, and it plays a very important role in building a European Research Area. It anticipates and complements the activities of the EU Framework

Programmes, and builds “bridges” towards the scientific communities of emerging countries. It also increases the mobility of researchers across Europe and fosters the establishment of scientific excellence in nine key domains. Our COST action FAIM is in the Food and Agriculture domain (www.cost.eu/about_cost).

Who is in our COST Action?

This unique COST Action (FAIM) brings together 120 to 200 experts from so far 20 (25) EU countries (and beyond). Number of participants is steadily growing. (www.cost.eu/domains_actions/fa/Actions/FA1102?management).

We started in late 2011 and will be “on the road” until 2015. The figure below shows our management structure.



What is our COST action FAIM about?

The title says it all, (almost!): “*Optimising and standardising non-destructive imaging and spectroscopic methods to improve the determination of body composition and meat quality in farm animals (FAIM).*” FAIM 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 (CCG) across countries. These actions are necessary for the development of value-based payment and marketing systems (VBMS) 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. This will be essential for achieving the required advances in CCG systems to measure carcass yield and MQ, to meet the industry need for VBMS, and to improve production efficiency throughout the meat supply chain (MSC). FAIM will also support EU legislation on individual animal identification through showing the additional benefits of feeding back abattoir data on individual animals for optimising management, breeding and providing phenotypic information which will facilitate future implementation of genome wide selection.

Our Objectives

- To review and develop robust references from imaging technologies for measuring body and carcass composition
- To review and develop harmonised procedures for in vivo, post-mortem and on-line imaging methods of predicting compositional traits
- To review and develop harmonised procedures for in vivo, post-mortem and on-line imaging and spectroscopic methods of predicting Meat Quality in livestock
- If full automation cannot be achieved, a lesser option is provided by semiautomatic methods, where results are obtained through human computer interaction
- To review and harmonise methods and equipment for individual animal traceability to optimise management, breeding and permit the future use of genomics.

Our means:

Annual Conferences (AC): first and latest (FAIM I) was in September 2012 in Dublin hosted by Teagasc Food Research Centre, Ashtown, Dublin. 24th - 26th September 2012. The second AC (FAIM II) will be in Kaposvár/Hungary (Kaposvár University; 29 & 30 of Oct.2013)

Workgroup meetings: mainly in connection with the AC but there is more: e.g. WG1 met in Jan. 2013 in Lyngby: Use of phantoms in computed tomography, e.g. WG 1 and WG2 will meet during the EAAP 2013 (26-30 Aug.) and FAIM will organise one session: Carcass and meat quality: from measurement to payment. e.g. WG3 will meet on “Farm Animal and Food Quality Imaging” in Espoo, Finland as satellite to Scandinavian Conference on Image Analysis (SCIA'13): 17/6/2013 and all WGs will meet at FAIM II in October

Training schools: we had 2 TS in 2012: (1) on image analysis in Lyngby, Denmark May 2012, (2) on Farm Animal Imaging & Carcass/Meat Quality in Oberschleissheim and Kulmbach Germany, October 2012.

STSMs (in full: Short term Scientific missions):

We had 6 STSMs in 2012 and we have the power to support more. Please come forward and ask!!

Where to find information about FAIM

Our Action website:

www.cost-faim.eu

FAIM website at main COST site:

www.cost.eu/domains_actions/fa/Actions/FA1102

Domain website:

www.cost.eu/domains_actions/fa

About this book

The papers included in this book are supplementary to the abstracts provided in the proceedings book received by delegates at the FAIM I conference held at Teagasc Food Research Centre, Ashtown, Dublin on the 25-26th September 2012. The papers in this book have not been peer reviewed.

Would you like to participate?

Email me! Lutz.Bunger@sruc.ac.uk



Participants of the FAIM training school in Kulmbach, Oct 2013



Participants of the FAIM training school in Oberschleissheim, Oct 2013

Overview to the FAIM I Meeting Farm Animal Imaging Opportunities and Challenges

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Value for industry

- Measurement is important for those meat producers and processors who wish to increase their business efficiencies and profitability.
- Imaging techniques offer meat producers and processors the opportunity to make a wide range of measurements on living animals, carcasses and meat in real time; including animal growth rate and health, body and carcass composition, and food safety and food quality.
- To get the benefits from taking measurements it is also important to have a good system for individual animal traceability which allows animal ID to be maintained from the farm through processing to packing.
- Science is taking up the challenge and offering solutions; but these need to be cost effective for industry to ensure up take.

Background

Humans have been creating images of animals since prehistoric times. Prehistoric paintings found in caves across Europe show clearly that early humans recorded images of the animals, and although there is considerable academic debate as to the purpose of the paintings, it might be suggested that the early humans were keen to record the shape and size of the animals that they were hunting.

With the growth and development of 'modern' agriculture, the recording of the size and shape of farm animals became more important, and more evident. In the nineteenth century, owners of prized livestock often commissioned paintings to show off the size and shape of their animals.

While size and shape remain important to livestock producers today, other factors such as the health, efficiency and ease of management of the animals are also important.

Modern farmers seek continually to improve both the quantity and quality of farm animals they produce. A key to sustainable improvement is measurement. Dr H James Harrington said:

“Measurement is the first step that leads to control and eventually to improvement. If you can't measure something, you can't understand it. If you can't understand it, you can't control it. If you can't control it, you can't improve it.”

Imaging technologies are already in use for making a number of measurements in farm animals, and have the major advantage over a number of other techniques in that they are non-destructive and in general non-invasive. So, if measurement is the key to control and thereby to improvement, which imaging technologies can be used for measurement, what are the future opportunities for imaging and where do challenges remain?

Current use of imaging

For breeders of farm animals, the production of viable offspring is essential. Here the use of ultrasound based imaging technologies has been established for a long time as a means of detecting pregnancy and predicting numbers of offspring to be born. More recently, the use of ultrasound techniques have allowed the detection of fetal movement which, in turn has been linked to the vigour of the newborn animal.

Figure 1. An image of a Steppe bison in the Altamira caves, Northern Spain painted more than 11,000 years ago in the Magdalenian period.



The growth rate of farm animals is an important measurement which often involves considerable time, effort and handling of livestock. The use of video image analysis of live animals, such as applied in groups of pigs, allows continuous non-invasive monitoring of the key dimensions of the animals. The data captured allows the estimation of the growth rate of groups of pigs, shows the level of variability within and between groups and give some indication of the overall health of the animals.

The health of farm animals can also be assessed to some extent using a direct imaging approach such as thermal imaging. The use of infrared thermography or thermal imaging can provide a means to measure temperature remotely and can be used to detect temperature in both the whole animal and in regions of the body.

Figure 2. An image of a hog at Tidmarsh Farm (c. 1798)
Source: English Museum of Rural Life



composition is important and a number of imaging based methods to assess body composition in live animals have been developed. Computed tomography or CT and Dual-Energy X-ray Absorptiometry or DEXA, use X-rays and image analysis, while magnetic resonance imaging uses magnetic and radio frequency fields and ultrasound scanning uses sound waves, to generate images which can be used to estimate body composition. There is a particular interest in the use of CT to replace some of the dissection based methods in pigs.

Imaging approaches, such as video image analysis, are also used to determine the composition and meat yield in carcasses. Indeed, video image analysis, is now quite widely used in the EU as a means of replacing the manual classification of beef carcasses using the EUROP grid.

Figure 3. An image of the “Ketton Ox” by R. Pollard, (1801)
Source: English Museum of Rural Life



Figure 4. Image depicting a Leicester Ram by R. Whitford, (1859)
Source: English Museum of Rural Life



Future Opportunities

The major benefit of imaging based approaches is that they are non-destructive, non-invasive measurements and allow remote monitoring so can offer a number of future opportunities for imaging in farm animal species.

With regard to food safety, imaging technologies may offer a low cost opportunity to food processors to monitor contamination. For example, the EU programme Prosafebeef explored the opportunities for the use of dietary markers and imaging techniques for the detection of faecal contamination on carcasses.

With regard to meat quality, recent developments have used a number of spectral techniques including near infrared spectroscopy, hyperspectral imaging, and raman spectroscopy to estimate the eating qualities of meat, particularly beef. Research progress in this area suggests that the prospect of being able to estimate eating and nutritional qualities of meat may not be too far away.

However there are also challenges ahead for farm animal imaging.

Future Challenges

Farm animal production and processing is both cost conscious and cost sensitive, so the costs of equipment and the labour required to operate it, represent major challenges.

For the estimation of body composition in live animals, the need for anaesthesia is a particular challenge, as is the size of the equipment is not large enough to accommodate large farm species. It is also important to assess a wide range of breeds and cross breeds to provide useful information for breeders and producers.

Estimation of live weight and growth rate needs to be carried out on an individual animal basis, so animal identity is important. A major challenge is to be able to use image based techniques in sheep where fleece growth and loss causes errors of estimation, and to be able to apply techniques equally well to animals kept outside as well as those housed inside.

In the assessment of carcass and meat quality, automation and integration into existing equipment and processes, which vary from business to business, is a very significant challenge.

Similarly, standardisation and validation of equipment and methodology throughout the EU will be an important challenge to address in the future. This is essential if these newer technologies are to show benefits for the farm animal production and processing industries.

Figure 5. A Charollais bull and calf (photo courtesy of Darren Todd, SRUC)



Body composition in farm animals by dual energy X-ray absorptiometry

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Value for industry

- Dual energy X-ray absorptiometry (DXA) non-invasively provides data for fat mass, soft tissue lean mass, bone mineral mass and bone mineral density in different farm animal species like pig, sheep, cattle (calves), poultry, and others for body weights up to 240 kg (in vivo or post mortem).
- New DXA machines provide rapid results and require minimum data analysis.
- The technology can be used for breeding or carcass classification purposes. Online measurements within the meat processing chain are possible.
- DXA compared with magnetic resonance imaging or computed tomography is very reasonably priced and provides a high output/cost ratio.

Background

The determination of body and carcass composition by dual energy X-ray absorptiometry (DXA) is based on the different X-ray attenuation coefficients (R value) of a low and of a high energy X-ray spectral level for soft tissue and bone mineral. Soft tissue consists of fat and lean tissue, which can be distinguished for tissue not overlying bone – also based on different X-ray attenuation coefficients (Pietrobelli *et al.*, 1996, Wang *et al.*, 2010, Stone and Turner 2012). The amount of fat within the soft tissue is linearly related with the R value. The amount of soft lean tissue and fat tissue overlying bone results from the composition of the bone neighbouring pixels by assuming for the bone containing pixels an identical soft tissue composition as in the non-bone containing neighbour pixels.

DXA provides a two-dimensional scan image of the whole body or regions of interest. A whole body or carcass scan image can be analysed totally or regionally by semi-automatically or manually defining regions of interest (Mitchell *et al.*, 2002).

Besides the amounts of soft lean or fat tissue and bone mineral content (each in g), DXA provides a measure of bone mineral density (g/cm²).

The time required for a whole body scan of a 120 kg pig varies between <3 and 40 minutes depending on device generation and/or software settings and decreases from pencil through fan to cone beam scanners. Smaller (shorter) animals (probes) take less time than larger ones, because the area to be scanned is reduced – as long as the scan settings stay unchanged. For scans in vivo, a sedation or anaesthesia of the farm animals is necessary in all cases. The whole body/carcass composition estimate is available immediately after the scan is finished and does not need further manipulation of the scan image. Alone, a regional analysis is a little time consuming depending on the number and anatomical specification of the regions of interest.

Why work is needed

Different generations of scanners offer a variety of solutions for the determination of bone mineralization and body/carcass composition measurements. Pencil beam scanners deliver a pixel-wise scan image, while fan and cone beam scanners deliver an area-wise scan image consisting also of separated pixels, either calculated by a software algorithm or measured directly by a linear or rectangular array of photon collecting sensors. The scan speed depends on the size and design of the photon collecting sensors, and different software or hardware settings like for example small animal, paediatric, or adult thin (quick), standard (normal), or thick (slow) modes. Therefore, DXA needs cross validation for transferring composition results among devices and software modes (Ruge 2006, Scholz *et al.*, 2007, Lösel *et al.*, 2010). Additionally, DXA as an indirect tool (Scholz and Mitchell, 2010) does not provide a measure of the lean meat percentage. It is necessary to determine the accuracy of DXA by reference dissection or chemical analysis.

The methods used

DXA has been applied on a variety of farm animal species e.g. chicken: Mitchell *et al.*, 1997, Swennen *et al.*, 2004, Schreiweis *et al.*, 2004, 2005; turkeys: Schöllhorn and Scholz 2007, Kreuzer 2008; pigs: Mitchell *et al.*, 1996a,b, 1998, Suster *et al.*, 2004, Hoffschulte and Scholz 2006, Bernau 2011, Kremer *et al.*, 2012; sheep: Rozeboom *et al.*, 1998, Scholz *et al.*, 2010,; and calves: Scholz *et al.*, 2003; Hampe *et al.*, 2005; Musick 2007 or beef: Mitchell *et al.*, 1997b, Ribeiro *et al.*, 2011; as well as in the wool and meat industry: Bartle *et al.*, 2004; Kröger *et al.*, 2005.

First studies dealt with the accuracy and precision of DXA to predict carcass (Svendsen *et al.*, 1993, Scholz *et al.*, 2002, 2010) and body composition (Scholz and Förster 2006, Musick 2007, Kreuzer 2008, Scholz *et al.*, 2010).

The following results are all based on studies performed at the Livestock Center Oberschleissheim using a GE Lunar DPX-IQ pencil beam scanner. The software modus “adult normal” was used for the in vivo and carcass swine studies, while the pediatric large modus was used for the in vivo calf and sheep study. Calf and lamb carcasses were studied with the pediatric small modus. The lamb carcass included both body sides without head, while only one carcass half without head was used for the calf study (Figure 1). All turkeys (whole body after euthanasia) were scanned by using the pediatric small modus. Dissection served as reference for pigs, lambs, and calves, while chemical analysis provided the reference values for turkeys.

The results obtained

Accuracy tends to be higher in pigs followed by poultry (turkey), sheep (lamb), and finally calves. Whole body analysis in sheep and calves in vivo is particularly strongly affected by the ruminant gastrointestinal tract leading to lower relationships between DXA body composition and reference measures (Tables 1 and 2). DXA is also able to discover (significant) differences in protein or energy among different treatments (e.g. breed, weight, food, gender) during growth (Mitchell and Scholz 2008).

Figure 1. Application of dual energy X-ray absorptiometry in farm animals (clockwise: calf, turkey body, calf carcass half, pig carcass half, lamb carcass, pig, sheep - in the middle DXA scan images (left: composite, right: soft tissue); all images from GE Lunar DPX-IQ or iDXA scanner in Oberschleisheim)

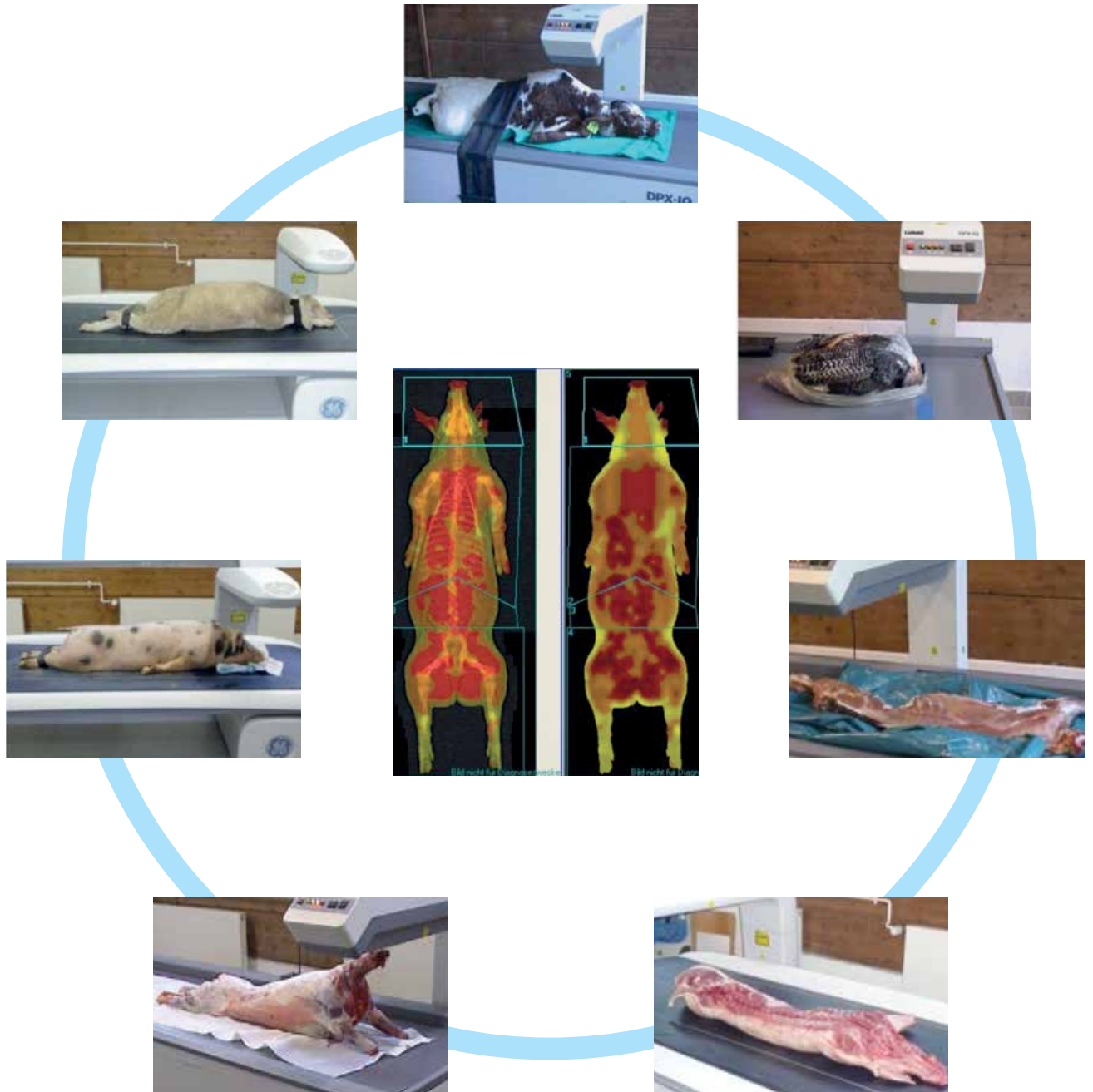


Table 1. DXA carcass vs. dissection or chemical analysis (alone Turkeys).

Dissection/chemical* reference vs. DXA carcass	Pig (n=61)	Lamb (n=93)	Calf (n=30)	Turkey* (n=100)
FAT %	R ² =0.80 √MSE=1.60	R ² =0.73 √MSE=1.68	R ² =0.28 √MSE=0.90	R ² =0.74 √MSE=2.11
FAT (g)	R ² =0.90 √MSE=359	R ² =0.83 √MSE=177	R ² =0.64 √MSE=179	R ² =0.86 √MSE=254
Meat or Lean* % /Soft Lean (%)	R ² =0.70 √MSE=1.89	R ² =0.57 √MSE=1.76	R ² =0.53 √MSE=1.95	R ² =0.69 √MSE=2.33
Meat or Lean* (g) /Soft Lean (g)	R ² =0.94 √MSE=848	R ² =0.88 √MSE=197	R ² =0.98 √MSE=329	R ² =0.99 √MSE=178
BM/Bone (%)	R ² =0.24 √MSE=0.64	R ² =0.03 √MSE=1.48	R ² =0.24 √MSE=2.38	R ² =0.01 √MSE=0.39
BMC/Bone (g)	R ² =0.73 √MSE=135	R ² =0.54 √MSE=127	R ² =0.77 √MSE=420	R ² =0.97 √MSE=27
Weight (g)	R ² =0.91 √MSE=696	R ² =0.94 √MSE=535	R ² =0.99 √MSE=295	R ² =0.99 √MSE=124

Table 2. DXA in vivo vs. dissection reference.

Dissection vs. DXA <i>in vivo</i>	Pig (n=61)	Lamb (n=93)	Calf (n=30)
FAT %	R ² =0.74 √MSE=1.72	R ² =0.51 √MSE=2.22	R ² =0.003 √MSE=1.06
FAT (g)	R ² =0.89 √MSE=969	R ² =0.71 √MSE=229	R ² =0.42 √MSE=228
Meat or Lean* % /Soft Lean (%)	R ² =0.65 √MSE=2.08	R ² =0.50 √MSE=1.88	R ² =0.09 √MSE=2.72
Meat or Lean* (g) /Soft Lean (g)	R ² =0.82 √MSE=2377	R ² =0.57 √MSE=369	R ² =0.94 √MSE=617
BM/Bone (%)	R ² =n.s √MSE=-	R ² =0.05 √MSE=1.53	R ² =0.26 √MSE=2.34
BMC/Bone (g)	R ² =0.73 √MSE=136	R ² =0.53 √MSE=129	R ² =0.84 √MSE=349
Weight (g)	R ² =0.91 √MSE=696	R ² =0.70 √MSE=1158	R ² =0.98 √MSE=1396

The scientific conclusions

DXA carcass analysis leads to a higher relationship with dissection or chemical analysis than DXA in vivo ($\Delta R^2 \geq 0.05$ for lean meat% or fat%; Scholz *et al.* 2002, 2007, 2010, Scholz and Förster 2006, Musick 2007, Kreuzer 2008, Tables 1 & 2: all data from the GE Lunar DPX IQ in Oberschleissheim). The prediction accuracy is even higher for tissue masses (e.g. in sheep: Mercier *et al.* 2006, Pearce *et al.* 2009) and for higher body weights or wide body weight ranges (Mitchell *et al.*, 1998, Mitchell and Scholz, 2009, Tables 1 & 2: all data from the GE Lunar DPX IQ in Oberschleissheim). Depending on the amount of fat in the carcass or in the body, and on the hardware or software settings, DXA may either overestimate or underestimate the amount of fat (lean meat) in comparison with the reference values from dissection or chemical analysis. Therefore species specific and/or even breed (genotype) as well as gender specific (regression) equations are necessary for an accurate prediction of the true body/carcass lean meat% or fat %.

The next steps

Based on the latest developments, DXA can move closer to MRI and CT, though it is still not possible to get three dimensional scan images for body composition analysis in one step. New rotating C arm devices are the first step towards three dimensional information. The advantage of very low radiation exposure with pencil and partially fan beam scanners, however, will disappear with three dimensional DXA. Devices combining DXA and CT technology are already available as so called DECT (dual energy computed tomography) devices. Research is needed to verify the possible applications of latest generations DXA scanners and DECT scanners for farm animal imaging in abattoirs or performance testing.

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Body composition of farm animals by MRI

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Value for industry

MRI can be used to:

- Predict body composition.
- Estimate carcass composition as an alternative to full dissection.
- Provide a carcass grading reference in performance testing.

Background

In livestock production and research there has always been a great demand for techniques to determine body composition of live animals and carcasses. Performance tests of meat producing animals, investigations of growth patterns and influencing factors are very much based on such techniques. In meat industry it is essential to evaluate carcasses in terms of lean meat and fat. Magnetic Resonance Imaging (MRI) is a development in medicine that has greatly enhanced the ability to visualize anatomical and pathological changes in vivo. MRI provides, without ionizing radiation, high contrast images of any desired plane.

The first whole body tomograph exclusively used for livestock science was installed in the Institute of Farm Animal Genetics in Mariensee in 1987 and was used until 2006. This paper reviews own research projects concerning predominantly the determination of body composition and the analysis of growth curves in swine, sheep, and water fowl.

The method used

MRI is a non invasive technique to acquire images of the body's interior in any desired plane. The basic principle is that atomic nuclei with an odd number of protons or neutrons or both will absorb and reemit radio waves when placed in a magnetic field. This phenomenon is called nuclear magnetic resonance (NMR) and has been widely used by chemists during the last 60 years. Hydrogen has the simplest nucleus, a single proton. It is most abundant in the body tissues, and its magnetic moment is large, both factors causing protons to emit a strong NMR signal.

This makes the hydrogen nucleus an attractive isotope for imaging. The MRI scanner used in the Institute of Farm Animal Genetics was a 'BRUKER Medspec BMT 15/100' whole body tomograph with a field strength of 1.5 Tesla. For acquisition of images with high contrast between muscle and fat tissue, a T1 weighted spin echo sequence was suitable. In a modified form, known as multi-slice multi-echo sequence, this method could generate adjacent slices with multiple echoes for each slice. The echoes provided information about the tissue specific relaxation of protons. By acquiring a set of parallel slices 3-D information of the body was delivered. The image matrix consisted of 256 rows and 256 columns. The field of view (FOV) was chosen according to the size of the animal and ranged from 260 x 260 mm to 460 x 460 mm, resulting in pixel sizes of 1.0 to 1.8 mm edge length. Slice thickness was set to 8 or 10 mm and slice distance to 16 or 20 mm. Depending on the length of the animal up to 90 parallel transverse images were necessary to cover the entire body. The measuring time for one set of 7 slices with 3 echoes each was about 4 minutes including image reconstruction. On average, one pig or sheep per hour could be scanned.

In medicine, image analysis is primarily a visual inspection to distinguish morphological and physiological alteration while in animal science; the main interest is quantification of body tissues. Simple measures are linear and area measurements, well known from other techniques utilized for livestock production such as ultrasound. Extensive information is available by means of multivariate pixel

classification like cluster analysis or Parzen window function. To prepare images for pixel classification, as a first step, regions of interest (ROI) were drawn to exclude those parts which did not contribute to lean and fat. The so masked images were analysed by the above mentioned classification methods. After multiplication by pixel size, the result obtained was muscle and fat area for each section or image. Applying the Cavalieri method volumes of muscle and fat tissue were estimated, not only for the whole body but also for its parts.

Animals have to be immobilized prior to scanning to avoid body movements. Motion artefacts can significantly reduce image quality. Animals have to breathe quietly but not very deeply, since strong breathing also causes motion artefacts.

The results obtained

In several experiments, methods for the estimation of body and carcass composition in pigs, sheep and water fowl were developed. Lean and fat content of German Landrace pigs of different weight groups (20, 50 and 90 kg live weight) were determined. Animals were scanned at five positions in the body: shoulder, breast, loin, sirloin and ham. Following tomography, pigs were slaughtered and carcasses dissected into lean, fat and bone as reference. MRI images were analysed by the image processing procedure, including cluster analysis, as described above. Table 1 shows the accuracy of estimation. It is obvious that MRI delivers a precise estimation of weight of total lean and fat for every weight group. The accuracy was very high for the percentage of lean and fat in the '90 kg' group, but reduced in the '20 kg' group. This might be due to the fact that the animals investigated showed a small variation in these traits and tissue differentiation was difficult (Baulain and Henning 2001).

Table 1. Accuracy of estimation of body composition in live pigs of different weight groups

	20 kg		50 kg		90 kg	
	R ²	SEE	R ²	SEE	R ²	SEE
Lean (g)	0.91	190	0.96	265	0.89	612
Fat (g)	0.89	90	0.97	150	0.91	374
Lean (%)	0.55	1.46	0.83	0.92	0.87	1.19
Fat (%)	0.68	1.06	0.80	0.97	0.89	1.01
	n = 43		n = 40		n = 60	

R²: Coefficient of determination; SEE: Standard error of estimation

Table 2. Accuracy of estimation of body composition in live lamb of different weight groups

	Live wt. ≤30 kg		Live wt. > 30 kg	
	R ²	SEE	R ²	SEE
Lean (g)	0.96	160	0.91	261
Fat (g)	0.96	84	0.94	195
Lean (%)	0.78	1.57	0.91	1.60
Fat (%)	0.86	1.49	0.90	1.64
	n = 49		n = 84	

R²: Coefficient of determination; SEE: Standard error of estimation

Different meat type lambs and their crosses with Finn sheep were scanned to derive equations for predicting body composition. One group of lambs weighed less than 30 kg. The second group was made up of lambs weighing more than 30 kg. Total dissection of the left carcass side into lean, fat

and bone served as reference. Prior to slaughter and dissection each sedated lamb was scanned at different regions of the body. The accuracy of estimation of body composition is indicated in Table 2. The coefficients of determination were at the same level as in the pig experiment (Streitz 1995).

Table 3. Correlations between muscle and fat volume in different water fowl species determined by means of MRI and total dissection

Species	n	Breast muscle	Leg muscle	Abdominal Fat
Peking duck	68/63/73	0.96	0.87	0.78
Muscovy duck	68/70/64	0.97	0.97	0.82
Mulard	78/77/73	0.98	0.84	0.84
Goose	72/70/64	0.96	0.80	0.84

A direct measurement of the breast and leg muscle volumes as well as abdominal fat volume was accomplished in water fowl. The volumes were determined by acquisition of adjacent transverse sections covering the entire length of the body. Applying the Cavalieri method it was possible to estimate the volumes with a high accuracy compared to the results of total dissection. Correlations between breast muscle volume and dissection weight, calculated between species and sex, ranged from $r = 0.95$ to 0.97 (Table 3) (Wiederhold 1996).

An essential requirement in studies on growth and

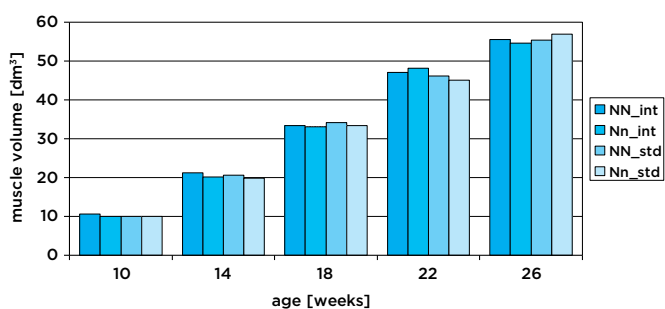
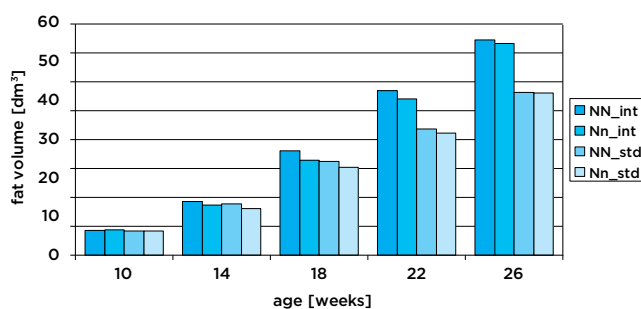


Figure 1: Muscle and fat growth of two MHS-genotypes (NN and Nn) in intensive and restricted fed pigs (n = 72)

development is a precise determination of body composition at different stages during the growth period. The most accurate method is manual carcass dissection into lean, fat and bone. To quantify tissue growth, stepwise slaughter of animals differing in age or body weight had to be carried out. But procedures which can be applied to animals are preferable. Based on cross-sectional MR images, tissue composition of growing pigs and lambs

were examined. From figure 1 it is evident that muscle growth of 72 intensively or restrictedly fed pigs of two malignant hyperthermia syndrome genotypes was not influenced by feeding system, while fat growth of intensively fed barrows was significantly higher in the finishing phase. Between MHS genotypes, no significant differences in tissue growth were found. Only in tendency, NN genotypes had a higher fat growth than Nn genotypes in the finishing phase (Kusec *et al.*, 2007).

Furthermore MRI proved to serve as a carcass grading reference in pig performance testing.



A total of 202 pigs originating from stationary sibling and progeny performance test were taken to estimate lean and fat in two commercial crossbred lines (Pi x Westhybrid and db.65 x db.classic), as well as purebred Piétrain (Pi), German Yorkshire (LW) and German Landrace (LR) pigs. Left carcass sides were scanned by MRI. Based on the series of images muscle and fat volumes of the whole carcass and virtual cuts were estimated. A full dissection of the

Table 4. Accuracy of carcass lean estimation in different pig breeds and crossbred types

	Piétrain	LW/LR	Pi*Westhybrid	db.65*db.classic
R ²	0.97	0.96	0.97	0.97
RMSE (kg)	0.43	0.46	0.59	0.62
CV	1.63	1.97	2.22	2.33
	n = 19	n = 36	n = 74	n = 73

R²: Coefficient of determination; RMSE: Root mean square error, CV = RMSE/mean

carcass sides according to the EU-method served as reference. The accuracy of muscle weight estimation is shown in Table 4 (Baulain *et al.*, 2010).

In addition to the traits regularly acquired in performance testing, carcass composition of 150 lambs was determined by MRI. Breeds were German Blackface, German Meat Merino, Leine Sheep, Bleu du Maine and Suffolk. Differences in carcass quality, based on conformation score and volumetric MRI, were ambiguous. Correlations between muscle volume measured by MRI and muscle scores ranged from 0.4 to 0.5 (Baulain *et al.*, 2011). MRI as a reference technique to estimate carcass composition can be applied instead of full dissection, when i.e. new measuring techniques or measuring sites have to be evaluated for its benefit in performance test.

In addition to the prediction of body composition, MRI can also be utilized to describe morphologic structures and pathologic changes. The quality of the images allows the identification of morphological abnormalities caused by particular housing conditions or diseases. An efficient use of MRI images for diagnostic purposes needs experience and guidance. For the anatomical orientation within the MR images and for the identification of organs and tissues, anatomical atlases are helpful (Figure 2) (Schulte Spechtel *et al.*, 1997).



Figure 2. MR images of a live turkey's breast (transverse and sagittal view)

The scientific conclusions

One of the main breeding goals in livestock production is the optimization of body composition under various genetic and environmental conditions. Consequently, there is a great demand for methods of determining tissue composition in live animals, carcasses and retail cuts. Medical imaging techniques are very suitable for this purpose. For field or on farm use robust and simple imaging equipment is essential, but advanced techniques are available for research. Ultrasound is most attractive for livestock production, where it has been used for several decades. MRI is another option as described above, but costs and complexity has limited its use.

The next steps

To date, MRI has been used mainly as a research tool, especially for determination of body composition and investigation of individual growth patterns. Future use should focus on diagnosis of production diseases, since current breeding goals include new traits of animal health and welfare. Furthermore, exact phenotypic measures of individuals are absolutely required for e.g. molecular genetic studies and characterization and evaluation of genetic resources.

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The use of computed tomography in small animal breeding

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Value for industry

- By means of computed tomography (CT) the body composition of the animals could be determined in vivo (on living animals).
- Using this technique in the selection of breeding animals, the dressing out percentage (meat production) of the offspring could be improved.
- By following changes in the body composition of the animals in vivo, the optimal slaughter age could be determined.
- By means of the CT, differences in the body composition of different breeds and sexes could be detected.
- Using this technique, the effect of different diets and/or other treatments on the volume and structure of different tissues could be detected.
- By the in vivo determination of egg yolk content, the hatchability of the eggs and the viability of the hatched birds could be improved.

Background

Computed tomography (CT) has been used for animal science since the early 1980s. In the past it was used for the in vivo determination of body composition and meat quality in various animal species. In this paper, the use of this technique in small animal breeding is reviewed.

Why work is needed

By reviewing the main fields and results of the use of computed tomography in small animal breeding, some recommendations will be given for its practical use.

The results obtained

In the case of chicken, computed tomography was mainly used for the determination of changes in the body composition during the rearing period (Bentsen and Sehested, Andrásy-Baka *et al.*, 2003). These experiments focused on the determination of the volume of the muscle and fat.

Beside following the changes in the body composition of the birds, differences in the body composition of different genotypes were also examined at given ages (Almási *et al.*, 2012). In these comparisons, three-dimensional histograms were sometimes used to demonstrate the differences between different body parts and/or at given anatomical points.

In special cases, three-dimensional reconstructions were also used for the comparison of the body composition of the different genotypes to demonstrate the differences in the volume and structure of different tissues (Figures 1-4).

Figure 1. Breast muscle of a 5-week-old Arbor Acres meat-type chick

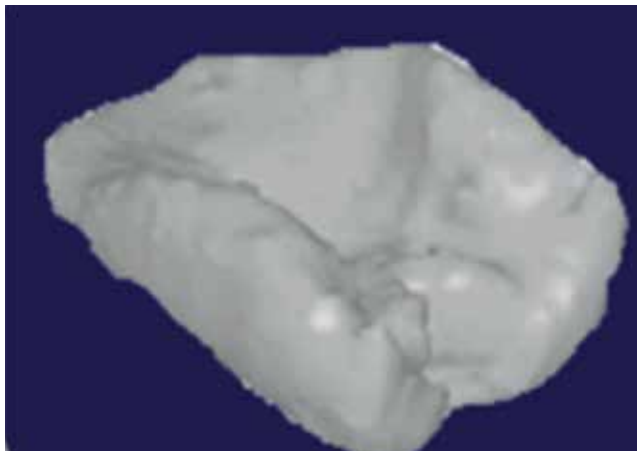


Figure 2. Breast muscle of a 20-week-old Tetra SL laying hen



Figure 3. Breast bone of a 5-week-old Arbor Acres meat-type chick

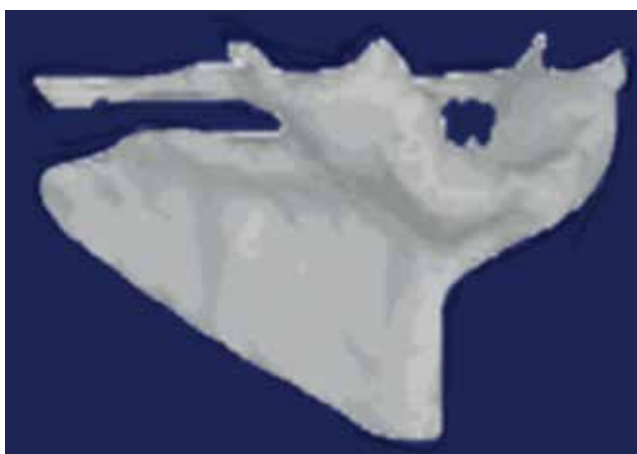


Figure 4. Breast bone of a 20-week-old Tetra SL laying hen



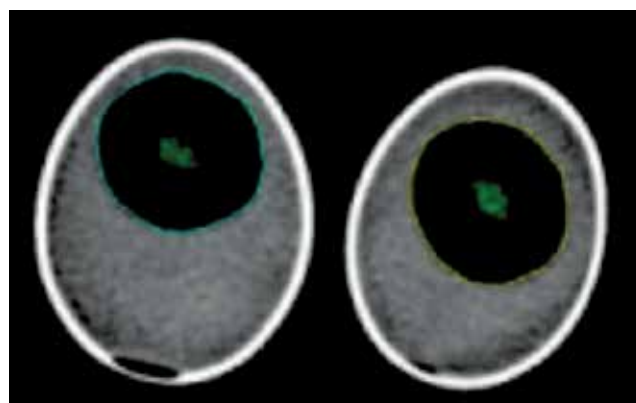
In laying hens, CT was used for following changes in the body fat content during the first egg laying period. In the experiment of Milisits *et al.*, (2010) it was pointed out that the body fat content increased till 44 weeks of age and it stagnated thereafter both in the brown and white egg layers.

Changes in the body composition of laying hens were monitored also during the moulting period by CT (Romvári *et al.*, 2005). In this study, the decrease in the amount of muscle and fat was demonstrated during the forced moulting period and their regeneration after a 3 week recovery period.

In connection with laying hens, CT was used also for the *in vivo* determination of the egg composition. However, in the study of Milisits *et al.*, (2009) it was established that the albumen and yolk are not separable based on their X-ray density values, because of their overlapping values on the Hounsfield-scale. The determination of the surface of

the yolk on the CT images (Figure 5) resulted in 70% accuracy in the prediction of egg yolk ratio.

Figure 5. Determination of the surface of egg yolk on cross-sectional CT images



Milisits *et al.*, (2010) observed that the body fat content of the hens and the yolk ratio of the eggs changed parallel during the first egg laying period. Between these two traits positive and significant correlations were obtained both in the brown and white egg layers ($r=0.401$ and $r=0.469$, respectively).

In the case of laying hens, CT was used also for the determination of the density, breaking strength and Ca content of the bones (Streubel *et al.*, 2005, Tossenberger *et al.*, 2011). The study of Tossenberger *et al.*, (2011) demonstrated how the diet's composition affects the X-ray density values (i.e. the composition) of the bones. The results of this experiment also pointed out that the Ca content of the bones can be predicted with about 60% accuracy based on their average Hounsfield values.

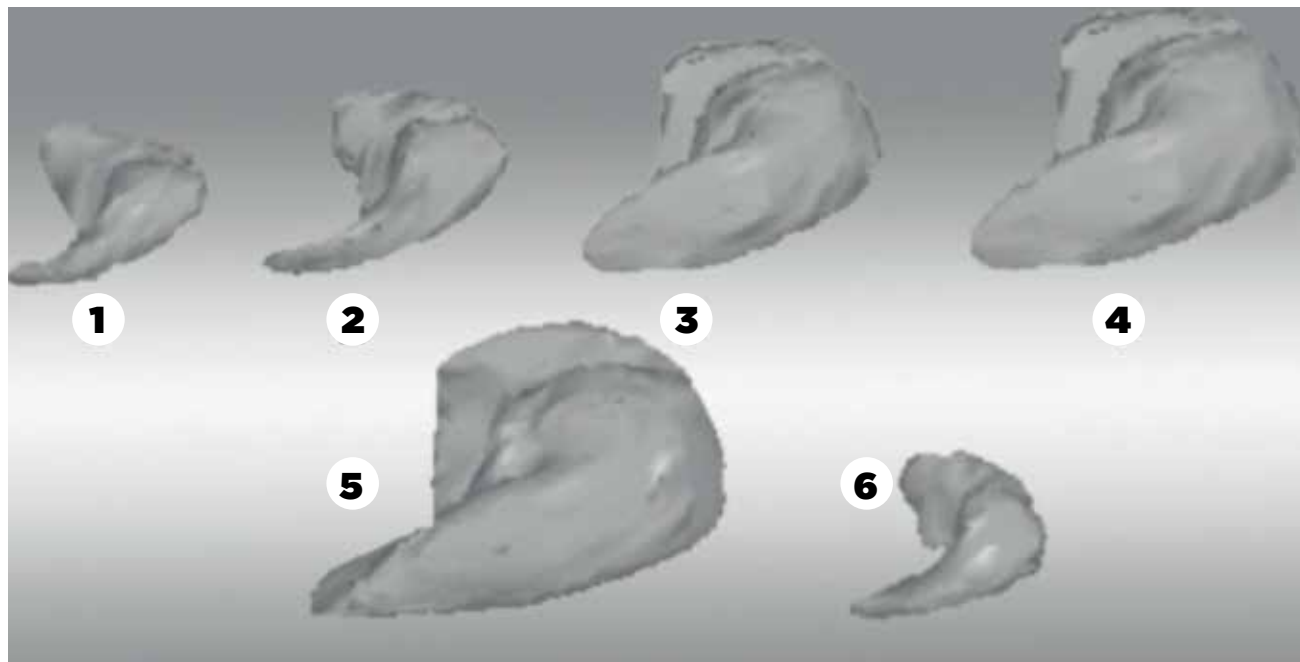
In the case of turkeys, CT was used also for following changes in the body composition of different genotypes during the rearing period (Brenoe and

Kolstad, 2000). Another interesting study in this species was carried out by Petneházy *et al.*, (2009) who determined the body composition and cardiovascular capacity of two different genotypes using CT and magnetic resonance imaging.

A special use of CT was undertaken for the preparation of a cross sectional anatomy atlas of the turkey (Petneházy *et al.*, 2012).

In the case of geese, CT was used for following the changes in the volume and composition of the liver during the force feeding period. In the study of Locsmándi *et al.*, (2005), three-dimensional reconstruction of the liver was used to demonstrate the increase in its volume during the force feeding period and its devolution thereafter (Figure 6). This experiment also pointed out, how the increased fat content of the liver affected the X-ray density values of this organ

Figure 6. Changes in the volume of goose liver during the rearing (1-2), force feeding (2-5) and devolution period (5-6) (1, 2, 3, 4, 5, 6 = 11, 15, 16, 17, 18 and 20 weeks of age, respectively).



In rabbits, CT was mainly used in the selection of breeding animals for improving the dressing out percentage of the offspring (Szendrő *et al.*, 1996). The selection was based first on the surface of *M. longissimus dorsi* and later also on the volume of the thigh muscle.

Using three-dimensional histograms, changes in the amount of body fat reserves during pregnancy and lactation were also demonstrated (Milisits *et al.*, 1999). This study also pointed out that the decrease in the body fat reserves of the does can be observed only in the last third of pregnancy.

A special use of CT was in the prediction of milk production of the rabbit does (Donkó *et al.*, 2008). In this case, the volume of the mammary gland was determined before and after nursing, while the milk production was predicted based on the calculated differences. The applied method seems to be suitable for estimating milk yield depending on the location of the pair of glands.

Computed tomography was also used for predicting the body composition of fish. In the study of Romvári *et al.* (2002) the body fat and protein content was predicted with high accuracy.

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The scientific conclusions

Based on the results, it was concluded that computed tomography seems to be a useful and efficient tool in a wide range of small animal breeding and production.

The next steps

The next step should be to broaden the use of this technique in the practice.

Use of computed tomography (CT) in a longitudinal body composition study in pigs fed different diets

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Value for industry

- Computed tomography (CT) is a non-invasive imaging technology, which can be used on live animals (in vivo) and on carcasses or primal cuts (post mortem).
- CT provides rapid, precise and accurate measurements or predictions of the weights of the three main tissues in meat animals: meat, fat and bone with accuracies (R^2 -values) of 0.99, 0.98 and 0.89, respectively or even higher when spiral CT is used.
- CT can also provide measurements such as; number of vertebrae which relates to the numbers of chops, the muscle density which is indicative for the intramuscular fat a major indicator of taste of meat and possibly for tenderness. CT also provides data on conformation and muscularity, bone density and pelvic dimensions, with the latter being probably indicative for dystocia and birth difficulties.
- CT scanning can provide the above described data on animals from a wide range of body weights (from mice and fish of about 30-50g, to chickens, rabbits, sheep and pigs with live weights of up to ca. 150 kg); the scanning process takes between 20 seconds up to 4 minutes.
- CT measured traits have moderate to high heritabilities permitting high selection responses and allow relaxation of the selection on CT traits to focus more on health and welfare traits. CT is a valuable tool as a benchmarking system and as integrated part of the breeding system.

Background

Although the main focus of CT based research work and commercial scanning at SRUC is on sheep (from 2000-2012 over 6000 lambs have been CT scanned), CT has also been used on other animals (from smaller model animals to various farm animal species) and its application in a longitudinal study on pigs will be described below as an example. This work was part of a larger project aiming to test the effects of low protein rations, with or without amino acid (AA) supplementation, on the performance and body composition changes of growing pigs of a lean commercial genotype (Bünger *et al.*, 2012). The main goals of this paper are to demonstrate in the FAIM context the use of CT in such longitudinal studies.

The European nitrate directive and increasing cost of protein sources are leading farmers to reduce the nitrogen content in livestock feed. UK pig production often employs feeds with relatively high protein levels to ensure high growth rates and low fat deposition, which are associated with undesired higher N-excretion. The aim of this work was to compare the performance of pigs of a lean genotype subjected to a conventional (C) feeding regime (FR) or one of two low protein FR (LP), supplemented with essential amino acids (AA) (LP1) or not (LP2). Performance was measured in terms of growth, feed intake, N-excretion/ N-retention and body composition with the latter measured repeatedly via CT and finally by slaughter.

Why work is needed

Research has shown that dietary protein can be reduced in the final stages of growth with only minor adverse effects on growth rate and feed conversion efficiency (Kerr and Easter, 1995; Le Bellego *et al.*, 2002), so long as dietary essential AA intakes and net energy (NE) are maintained. However, at the lowest levels of protein, a tendency to increased fatness has been observed (e.g. Canh *et al.*, 1998; Kerr and Easter, 1995). The effects of low protein diets are expected to be greater the leaner the genotype (Wood *et al.*, 2004).

The pattern of fat deposition in finishing pigs is important (Wood, 1984; Kouba *et al.*, 1999; Kouba and Sellier, 2011). Fat deposited in subcutaneous depots is unwanted, leading to increased requirements for fat trimming at the abattoir and a reduction in the price paid to the producer. However, intramuscular fat (IMF) has potential sensory benefits for meat quality, so deposition of fat within the muscle could enhance product quality (Teye *et al.*, 2006). Fat deposition can also occur within the body cavity, around internal organs.

Although this has little influence on carcass quality, deposition of internal fat does affect the efficiency of growth and meat production. Information about partitioning of fat between body depots is usually gained from dissection studies, which are time-consuming and expensive, as meat cannot be returned to the food chain.

The methods used

CT scanning has the ability to describe and follow the changes in whole body composition across time in live animals, in a non-invasive and non-destructive manner (Bünger *et al.*, 2011). This imaging technique, and associated image analysis methods, can also identify and quantify fat in different depots. CT research in sheep has shown that fat in different carcass and internal depots can be accurately quantified in ewes (Lambe *et al.*, 2003) and lambs (Lambe *et al.*, 2006; Young *et al.*, 1996) using information from a small number (3-5) of cross-sectional reference scans taken at set anatomical positions along the length of the body in prediction equations. However, breed or line specific calibration trials are required, to relate reference scan data to dissected tissue weights, to derive these prediction equations, and such equations become less reliable as genotypes change due to selection. This is a greater issue in pig breeding, where a faster rate of genetic progress is achieved compared to sheep breeding. Alternatively, many cross-sectional CT scans can be taken at regular intervals (usually at 8 mm distance) along the length of the body and total volumes of different body tissues can then be estimated (Roberts *et al.*, 1993). Using tissue density, tissue volumes can then be transformed into very accurate tissue weights.

Pigs were weighed on arrival and allocated randomly to pens and treatments, so that the average live weight on each treatment was as similar as possible. To enable growth rates to be calculated, all pigs were weighed weekly. Pigs were CT scanned three times, at an average weight of 60kg (scan 1), 85kg (scan 2) and 115kg (scan 3), following administration of a general anaesthetic, to minimise stress and movement during the scanning process. Food was withdrawn overnight prior to CT (for < 24h) to reduce gut fill. The study found no effect of CT scanning, including food withdrawal, on growth rate and feed intake. All procedures involving animals were approved by the SRUC animal ethics committee and were performed under UK Home Office licence, following the regulations of the Animals (Scientific Procedures) Act 1986. Altogether, the pigs were on the experiment for 73 to 88 days, depending on batch.

Figure 1. Application of CT scanning in pigs (experimental pigs before scanning; the CT scanner at SRUC (Siemens, SOMATOM Esprit); anaesthetized pig in CT scanner, examples for cross sectional images of which for example from this pig 155 were taken; 3D reconstruction of the carcass relevant parts of the pig using all 155 images, changing the threshold makes the skeleton visible, the pig waking up from anaesthesia).



The results obtained

Pigs on LP2 had lower LW and average daily weight gain (ADG) at key stages and higher feed conversion ratio (FCR) than pigs on C or LP1 from the point where all pigs reached an average of 60 kg LW onwards. The growth curves for C and LP1 animals were very similar to each other, and overall the results confirm that the LP1 strategy allows growth performance similar to the C FR. The N-intake during the trial was 11% lower in LP1 than C and 16% lower in LP2. CT scan data, which enabled the calculation of N-retention and excretion over the 60 to 115 kg period, showed that the LP1 FR can reduce N-excretion by about 17% in comparison with FR C, without compromising growth performance, however FCR was significantly poorer (-6%) in LP1 compared to FR C.

In vivo CT measurements agreed well with sample joint (fore loin) dissection results after slaughter for the main carcass tissue weights (fat $r = 0.88$; muscle $r = 0.71$; bone $r = 0.48$) and proportions (fat% $r = 0.92$; muscle% $r = 0.79$; bone% $r = 0.32$

with the lowest correlations for the bone traits. CT-measured muscle density was a good predictor of intramuscular fat ($r = 0.71$). Pigs on the C and LP1 diets did not differ significantly in composition during growth. However, pigs on the LP2 diet had significantly more fat (in carcass, internal and intra-muscular depots) and less muscle, from 85 kg onwards. Although total fat levels differed depending on diet (LP2 > others), proportions of fat in different body depots were not affected.

The slaughter results confirmed that FR LP2 produced fatter pigs in terms of subcutaneous, intermuscular and intramuscular fat (IMF), the latter measured in *longissimus* and *semimembranosus*. The percentage of 18:2n-6 fatty acids was lower and that of 18:1n-9 was higher in IMF from LP2 pigs. Pigs in FR LP1 had more *longissimus* IMF than C, with a different fatty acid composition but total fat deposition was similar in these 2 groups. The high IMF content in LP2 produced more tender, juicy steaks.

The scientific conclusions

• Nitrogen excretion/retention

A reduction of nitrogen excretion was successfully achieved by supplying the pigs with low protein rations, however only LP1 had an N retention comparable with the C group. Substantially lower excretion values on the LP1 diet can be achieved without compromising the growth performance; but, looking at the whole trial period, the feed required for an LP1 animal to produce a LW gain of 1kg was 6% more than that required for a C pig. Given however, that FCR has a moderate heritability, and considering that these pigs are probably adapted to C-like diets, there seems to be an opportunity for the use of genetic selection to counterbalance the decrease in efficiency on LP1.

CT can give accurate estimates of tissue proportions and distribution across the growing period in individual animals

- Fewer animals required in growth studies compared to serial dissection
- Higher accuracy than serial dissection – same animals followed through growth
- Prediction of important meat quality indicators in vivo is possible

• In vivo CT predictions of IMF

Most note-worthy is the association between IMF and CT muscle density in live pigs, as has been found previously in sheep in other SRUC research. Correlations of around 0.7 suggest that CT can predict this important meat quality trait in vivo with good accuracy. Very few accurate in vivo predictors of meat quality are available, so this result is of great interest. Despite a low coefficient of variation for CT-measured muscle density (< 3% at each scan), significant differences could still be detected between treatment groups in these pigs where

IMF levels were very low across the population (mean 1.2%, range 0.6 to 2.7%). These results suggest great potential for CT to provide both valuable carcass composition and meat quality data for live selection candidates within pig breeding programmes.

The increases IMF, in pigs fed the LP2 diet at the 85 kg and 115 kg scans, imply that IMF levels can be manipulated by altering protein and amino acid levels in the feed. This would give potential to increase eating quality of the meat, but this would be associated with an increase in fat in other, unwanted, depots. A more targeted approach to maintaining or increasing IMF, whilst reducing fat in other carcass and internal depots, would be through genetic or genomic selection. Although genetic correlations between IMF and other fat depots, such as subcutaneous fat, are positive, they tend to be only moderate in size in pigs, indicating a potential to select for fat in one depot and against fat in another. However, whilst payments to producers are based on the current policy, which penalises subcutaneous fat and does not reward meat quality, incentives for adopting such breeding strategies are low.

The next steps

- Further studies should aim to investigate the best predictor or combination of predictors from spiral CT scan images information of intramuscular fat percentage
- Optimised predictors should be measured on a powerful pedigreed sample of pigs to estimate robust genetic parameters and then incorporated into a breeding program

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Overview of the technical characteristics of systems predicting carcass, meat eating and nutritional quality of meat

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Value for industry

- Ultrasound and live animal video image analysis (VIA) methods can contribute to more accurate prediction measures of carcass quality.
- Carcass VIA systems are now used commercially and offer additional advantages in providing yield information as well as classification.
- Objective tenderness measures associate with trained sensory panel scores and provide a cost-effective and quick method for assessing (arguably) the most important quality parameter.
- Spectroscopic systems that provide estimates of meat sensory and nutritional quality, can be deployed in the abattoir or meat processor and operate on-line, and at line speed.

Background

It is possible to perform a range of measurements on both live animals and carcass/primary meat products to assess carcass, meat eating and nutritional quality. Some of these are feasible to be implemented in a commercial on-line environment and show promise in estimating the range of quality parameters that could relate to, and inform, the value chain. A range of techniques employed to assess meat and carcass quality, are herein described, and are capable of use in an on-line or commercial environment.

Why the work is needed

In response to industry demand, a number of options have been tested for performance and capability for measurement of meat and carcass quality in our recent studies. These include measures from both carcasses and live animals to obtain yield and quality measures. Details on the technical specifications, if not outlined in this paper, are contained in the associated paper references.

Methods and results

On the live animal, techniques such as imaging ultrasound to estimate intra-muscular fat and carcass yield have been developed. Lambe *et al.*, (2010) showed that measurement of fat and muscle depths by extracting dimensions from acquired ultrasound images taken at the start and end of a finishing period, in combination with live-weights, provided good estimations of carcass muscle and fat yields ($R^2 = 0.81$ and 0.84 , respectively). Image analysis techniques applied to serial sequences of ultrasound images have revealed reasonable relationships with intra-muscular fat content ($R^2=0.48$), even at low levels of intramuscular fat (Glasbey *et al.*, 2012).

Imaging systems have been applied to live animals to estimate both live-weight, and predict carcass classification and yield parameters of beef animals with some success. A recent study showed that carcass weight could be estimated accurately with an R^2 of 0.93 (Hyslop *et al.*, 2009).

Images of top, profile and rear of animal were gathered automatically in commercial conditions. Other carcass measures could also be predicted with reasonable precision, such as sirloin weight ($R^2 = 0.85$). Carcass-based video image analysis (VIA) systems are now commercially applied in various territories and offer advantages in prediction of yield, as well as carcass classification. These systems gather images of carcasses on-line, and some use structured light to interpolate three-dimensional morphology. A number of trials have been conducted to assess the performance of these systems. VIA machines, trial references and performances obtained are summarised in tables 1 and 2 below.

Table 1. VIA machine models and manufacturers

VIA machine reference	Manufacturer
BCC2	Carometec A/S, Denmark
VBS2000	E+V GmbH, Germany
MAC	Normaclass, France
VIAScan	Cedar Creek, Australia
CVS whole carcass camera system	RMS USA

Table 2. VIA reported performance studies

Trial	Machine designation	Performance
Sorensson <i>et al.</i> , (1988)	BCC1	LMY $R^2 = 0.73$, fat $R^2 = 0.77$, bone $R^2=0.79$
Eldridge (1994), Ferguson <i>et al.</i> , (1995)	VIAScan	Meat yield (kg) $R^2 = 0.98$ (with fat depth measure)
DMRI (1996)	BCC2	Conformation $R^2 = 0.90$; fat $R^2 = 0.63$
Sonnichsen <i>et al.</i> , (1998)	VBS2000	Conformation $R^2 = 0.91$; fat $R^2 = 0.83$
Lebert <i>et al.</i> , (2000)	MAC	Conformation $R^2 = 0.93$; fat $R^2 = 0.68$
Allen <i>et al.</i> , (2000)	Multi-machine	Within 1 sub-class $R^2 = 0.91 - 0.97$; fat $R^2 = 0.72 - 0.80$

In addition to the details outlined above, Craigie *et al.*, (2010) identified high value primal predictions using VIA input variables, not to be significantly better than grader. However, Pabiou *et al.*, (2011) found good associations between VIA data and low, medium and high value cuts. For example, high value cut weight predictions were considered very good ($R^2 = 0.93$). All the above studies relate to cattle. A VIA study in sheep, reported by Rius-Vilarrasa *et al.*, (2009; 2010), showed better precision than the grader for predicting primal cuts (86-99% accuracy), with very good repeatability (up to 99%).

Ross *et al.*, (2009) showed that objective measurements relating to tenderness could estimate the prediction of scientific sensory panel scores. For some types of measure, the prediction was enhanced by assessment at 72 hours post-mortem compared with at 14 day maturation, with correlation coefficients of 0.6 and 0.47 respectively.

Commercial exploitation of these types of techniques have been facilitated by the introduction of an industry-deployable robust and portable, multi-test instrument the 'Tenderscot' (www.pentlandprecision.co.uk/page.php?id=99). This instrument is set up to carry out Warner-Bratzler, MIRINZ bite and Rapid Slice Shear tests in alternative configurations.

It is possible to deploy certain spectroscopic measurement systems, such as visible and near infra-red reflectance spectroscopic surface probes, in the processing chain to estimate a range of sensory and nutritional characteristics. These systems can include measuring the reflected optical spectrum of a controlled, illuminated meat surface via a fibre optic guide. Results of trials on these systems are reported in another paper (Roehle *et al.*,) published in this same proceedings.

The scientific conclusions and next steps

There are a range of technological systems that can usefully inform on carcass and meat quality, throughout the production chain from farm to the abattoir (post-mortem). Studies to validate these systems in commercial conditions are now needed to show robustness of performance in different commercial settings.

The Quality Meat Scotland and Scottish Government funded Integrated Measurement of Eating Quality project aims to assess the viability of a range of these techniques when applied to a robust, on-line environment. These techniques

include autonomous, robotic-based probe systems, capturing data on meat and carcass quality parameters. These systems have been recently demonstrated to operate and gather data at fast commercial rates. (Figure 1 right), and are therefore suitable for future development and refinement to fully automated commercial systems.

A range of alternative spectroscopic systems for non-destructive evaluation of meat sensory and nutritional properties are being tested and are showing promise for on-line implementation. However, it is likely that there will be a need for further refinement, and robustness testing in a range of commercial environments.



Figure 1. Autonomous robotic arm concept applying probes on-line to carcass line

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Investigations into relationships between visible-near infrared (NIR) spectra and instrumental meat quality parameters in lamb *M. longissimus lumborum* and *M. semimembranosus*

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Value for industry

- Visible-near infrared (NIR) spectra from lamb *M. longissimus lumborum* (LL) showed some promise for predicting intramuscular fat percentage in that muscle.
- NIR spectra collected on the LL had a limited ability to predict Volodkevich shear force ($RPD_{pred} = 1.17$) or MIRINZ shear force ($RPD_{pred} = 1.33$). Models were unable to correctly identify all samples with shear force values > 5.5 kgF.
- Shear force traits in *M. semimembranosus* (SM) could not be predicted by NIR spectra collected on the LL.
- NIR was unable to predict pH_{ult} of LL or SM.
- These results were obtained on experimental lambs under controlled experimental conditions; future experiments should investigate NIR performance under commercial operating conditions.
- This research showed that cross validation methods underestimate the standard error of prediction for all traits.

Background

The ability to measure some meat quality traits as lambs are processed would give processors the ability to supply customers on a meat quality basis and to reward farmers who produce superior lambs with superior meat quality characteristics. Meat quality information, if made available to sheep breeders, would allow breed improvement strategies to select for meat quality traits aligned to customer requirements.

The ability to measure safely lamb meat quality in a non-destructive and cost-effective way is an

important prerequisite of any system intended for industry application. Visible-near infrared spectroscopy (NIR) meets these criteria, and has shown some promise for predicting beef and pork meat quality, while the application to lamb meat quality has been less well investigated.

The aim of this experiment was to determine the ability of NIR spectroscopy data collected on fresh (never-frozen) lamb LL to predict instrumental meat quality parameters of *M. longissimus lumborum* and *M. semimembranosus*.

Materials and methods

A total of 208 Texel lambs were slaughtered at a mean age of 144 days (range 126 to 155 days) and mean hot carcass weight (HCW) of 15.1 kg (range 8 to 25 kg) on a single day in a commercial abattoir and were subjected to electrical stimulation (825 volts, 14 Hertz for 20 seconds) at approximately 40 minutes post mortem.

The methods for meat quality measurements are described by Craigie *et al.*, (2012), briefly, traits measured on the LL included Volodkevitch and MIRINZ shear force, intramuscular fat percentage (IMF%), moisture content (%) and ultimate pH. Traits on the SM included Warner-Bratzler shear force, colour (L^* , a^* , b^* , chroma and hue), ultimate pH and cooking loss expressed as a percentage of the un-cooked weight.

After the LL was removed from the carcass at 7, 8 or 9 days post mortem, a 15 mm slice was taken from the anterior end of the muscle for NIR spectra collection. The freshly cut surface was allowed to bloom for two minutes (Shackelford *et al.*, 2005). An ASD Labspec 5000 (ASD Inc., Boulder Colorado) NIR spectrometer fitted with a high-intensity contact probe (Figure 1) with a 10 mm spot size was operated using a laptop computer running the Indico Pro program (ASD Inc.). Ten replicate NIR spectra (350–2500 nm at 1 nm intervals) were collected by removing and replacing the scanning head on the meat surface between scans.

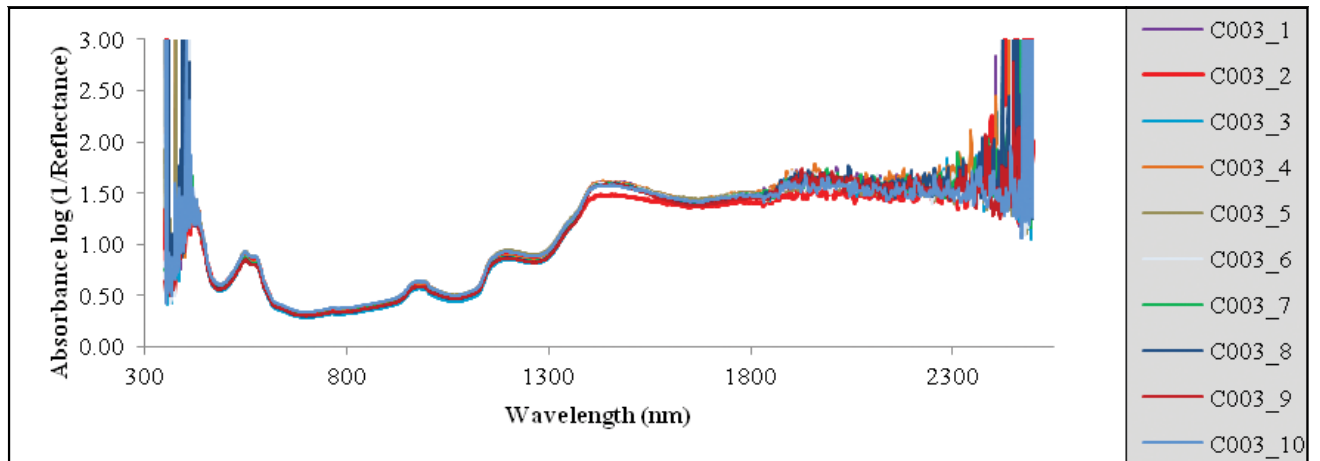
Spectra were recorded as absorbance log (1/Reflectance). Plotting all spectra revealed that regions at the extremes of the range (350–2500 nm) contained excessive noise (Figure 2). Removing these sections (350 to 499 nm and 1801–2500 nm) resulted in 500–1800 nm as the working spectra.

Samples were split into calibration and prediction datasets by sorting in ascending order separately for each parameter and selecting every fourth sample for the prediction dataset, with the intervening three samples being allocated to the calibration dataset as recommended by Williams (2001). Partial least squares regression type 1 was used for predicting instrumental meat quality traits on the three muscles using median NIR spectra (500–1800 nm) from the replicates that had not been rejected as outliers as explanatory variables. Westerhaus *et al.*, (2004) describe a strategy for handling outliers. Following this strategy, outliers for the reference meat quality parameter were identified when the calibration, cross-validation or prediction performance was poor, however samples were only removed if there was a known error with the sample value or where measurements were > 3 SD from the mean of the dataset. Potential outlier spectra were first identified in the same way, through poor calibration, cross validation or prediction performances. The Hotelling T^2 statistic is a generalization of the Student's t-test for multivariate analysis (Hotelling, 1931).

Figure 1. The ASD Labspec 5000 NIR spectrometer (left), the high intensity contact probe (centre) and application to a slice of lamb *M. longissimus lumborum*



Figure 2. Ten replicate scans from one sample (C003) over the full range (350-2500 nm), excessive noise can be seen at the ends of the spectral region.



Results

After the exclusion of an extreme value (10.68 kgF) in the MIRINZ prediction dataset, the R^2_{pred} was the same as the $R^2_{\text{pred}} = 44\%$ reported by McGlone *et al.*, (2005) who applied their model to predict MIRINZ shear force on an independent dataset of 12 lambs with unknown backgrounds. Retaining the outlier reduced the R^2_{pred} to 36%, $SE_{\text{pred}} = 1.38$ (data not shown). In the prediction dataset for Volodkevitch shear force, there were three samples that had shear force values > 5.5 kgF which has been identified as a toughness threshold above which there may be adverse consumer reaction (Lambe *et al.*, 2011). The model was unable to identify these samples as having a shear force value > 5.5 kgF (results not shown). In the MIRINZ prediction dataset, four samples that had MIRINZ shear force values > 5.5 kgF, despite the encouraging performance, the model only correctly predicted one out of four samples that had a MIRINZ shear force value > 5.5 kgF, although of the remaining 38 samples with reference MIRINZ shear force values < 5.5 kgF, none were predicted above this value (results not shown).

After removal of two outliers, a high R^2_{cal} was obtained for intramuscular fat percentage (IMF%) ($R^2_{\text{cal}} = 69\%$, $SE_{\text{cv}} = 0.36\%$), and the prediction performance was similar ($R^2_{\text{pred}} = 65\%$, $SE_{\text{pred}} = 0.33$, $RPD_{\text{pred}} = 1.68$). There was a marked decline in prediction of moisture content and ultimate pH. NIR was poor at predicting the shear force of the SM (Table 2) which was expected given that Volodkevitch shear force was found to be poorly correlated between the LL and the *M. vastus lateralis* in these lambs (Lambe *et al.*, 2011). A correlation between LL and SM could not be established because two different shear force tests were used. NIR did show some promise for predicting the chroma of lamb SM, $R^2_{\text{pred}} = 57\%$ ($SE_{\text{pred}} = 1.03$, $RPD_{\text{pred}} = 1.55$), however the performance was not as good for predicting the hue ($RPD_{\text{pred}} = 1.19$). RPD_{pred} values of 1.57 for L^* , 1.47 for a^* and 1.26 for b^* were obtained when predicting SM colour from NIR spectra collected on the LL.

Table 1. Performance of NIR calibration equations showing the coefficient of determination (R^2) and standard error (SE) for calibration and cross-validation for predicting instrumental meat quality parameters of lamb from spectra collected on *M. longissimus lumborum*. The calibration phase was performed on 70% of the data.

			Calibration			Cross-validation		
Lamb meat quality parameter	Pretreatment ^a	PC ^b	n ^c	R ² (%)	RMSE ^d	R ² (%)	SE	RPD ^e
<i>M. longissimus lumborum</i>								
Volodkevich shear force (kgF)	Baseline	2	154	24.6	1.03	21.4	1.06	1.31
MIRINZ shear force (kgF)	Baseline	2	126	24.6	1.27	18.7	1.34	1.10
Intramuscular fat (%)	Baseline	7	151	68.5	0.31	57.4	0.36	1.88
Moisture content (%)	MSC	3	152	38.1	0.71	32.6	0.75	1.33
Ultimate pH (pH _{ult})	Baseline	5	96	31.7	0.08	13.3	0.09	1.37
<i>M. semimembranosus</i>								
Warner-Bratzler shear force (N)	na	0	156	0	na ^f	na ^f	na ^f	na ^f
Lightness (L*)	none	3	156	49.7	1.53	46.7	1.59	1.36
Redness (a*)	none	6	156	56.7	0.85	46.7	0.95	1.36
Yellowness (b*)	MSC	6	153	49.1	0.80	30.0	0.95	1.20
Chroma (C*)	MSC	7	156	59.8	0.95	37.8	1.19	1.25
Hue (H*)	Baseline	4	156	24.1	0.03	12.2	0.03	1.06
Ultimate pH (pH _{ult})	MSC	0	156	0	na ^f	na ^f	na ^f	na ^f
Cooking loss (%)	SNV	3	153	37.6	2.05	32.7	2.15	1.21

^a Pre-treatments applied to the spectra prior to PLS regression analysis and prediction,

Baseline = Baseline correction,

MSC = Multiplicative Scatter Correction and SNV = Standard Normal Variate.

^bPC = number of principal components used in the regression.

^cn = number of samples included in calibration and cross-validation phases.

^dRMSE = root mean square error.

^eRPD = ratio performance deviation is the SD of the Y variable in the calibration dataset (after removal of outliers) divided by

^fna = not available, where the calibration, cross-validation or prediction phase failed.

Table 2. Performance of NIR calibration equations showing the coefficient of determination (R^2) and standard error (SE) for prediction of instrumental meat quality parameters of lamb from spectra collected on *M. longissimus lumborum*. The prediction phase was performed on 30% of the data to gauge predictive ability.

Lamb meat quality parameter	Pretreatment ^a	PC ^b	n ^c	Prediction		
				R ² (%)	SE ^d	RPD ^d
<i>M. longissimus lumborum</i>						
Volodkevich shear force (kgF)	Baseline	2	52	29.1	1.05	1.17
MIRINZ shear force (kgF)	Baseline	2	42	44.0	1.04	1.33
Intramuscular fat (%)	Baseline	7	51	64.9	0.33	1.68
Moisture content (%)	MSC	3	52	8.7	0.91	1.06
Ultimate pH (pH _{ult})	Baseline	5	33	0	0.10	1.00
<i>M. semimembranosus</i>						
Warner-Bratzler shear force (N)	na	0	51	na ^e	na ^e	na ^e
Lightness (L*)	none	3	52	59.7	1.34	1.57
Redness (a*)	none	6	52	53.9	0.92	1.47
Yellowness (b*)	MSC	6	51	36.0	0.96	1.26
Chroma (C*)	MSC	7	52	57.4	1.03	1.55
Hue (H*)	Baseline	4	52	29.3	0.03	1.19
Ultimate pH (pH _{ult})	MSC	0	51	na ^e	na ^e	na ^e
Cooking loss (%)	SNV	3	51	29.5	2.11	1.21

^a Pre-treatments applied to the spectra prior to PLS regression analysis and prediction, Baseline = Baseline correction, MSC = Multiplicative Scatter Correction and SNV = Standard Normal Variate.

^b PC = number of principal components used in the regression.

^c n = number of samples used for the prediction phase.

^d RPD = ratio of performance deviation (SD of the prediction dataset divided by the SE of prediction).

^e na = not available, where the calibration, cross-validation or prediction phase failed.

The scientific conclusions and next steps

This research showed that it is possible to predict some lamb meat quality parameters with NIR spectroscopy under experimental conditions. Development of stable prediction equations is dependent on having adequate variation in the trait of interest and a sufficiently large data set. Although one of the largest NIR data sets analysed to date in lamb meat, the lambs were part of a scientific trial and consequently should not be considered representative of a commercial slaughter population. For example, the vast majority of lambs slaughtered in the UK are crossbreds and the lambs used in this experiment were a specific breed type and there was very little variation in pH and shear force traits.

The current results showed that there are differences in traits between muscles and that spectra collected on one muscle may not necessarily be a good predictor of meat quality traits of traits in another muscle. Further research is required to establish the robustness of the prediction equations and to further refine NIR calibration equations to measure lamb meat quality before NIR can be recommended as a tool ready for implementation. Next steps should aim to develop calibration models that are applicable to a wider variety of muscle and breed types in a number of different abattoirs and to investigate novel analysis techniques for development of such models.

Acknowledgements

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Identification of the most appropriate improved measurement techniques for predicting carcass, meat eating and nutritional quality

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Value for industry

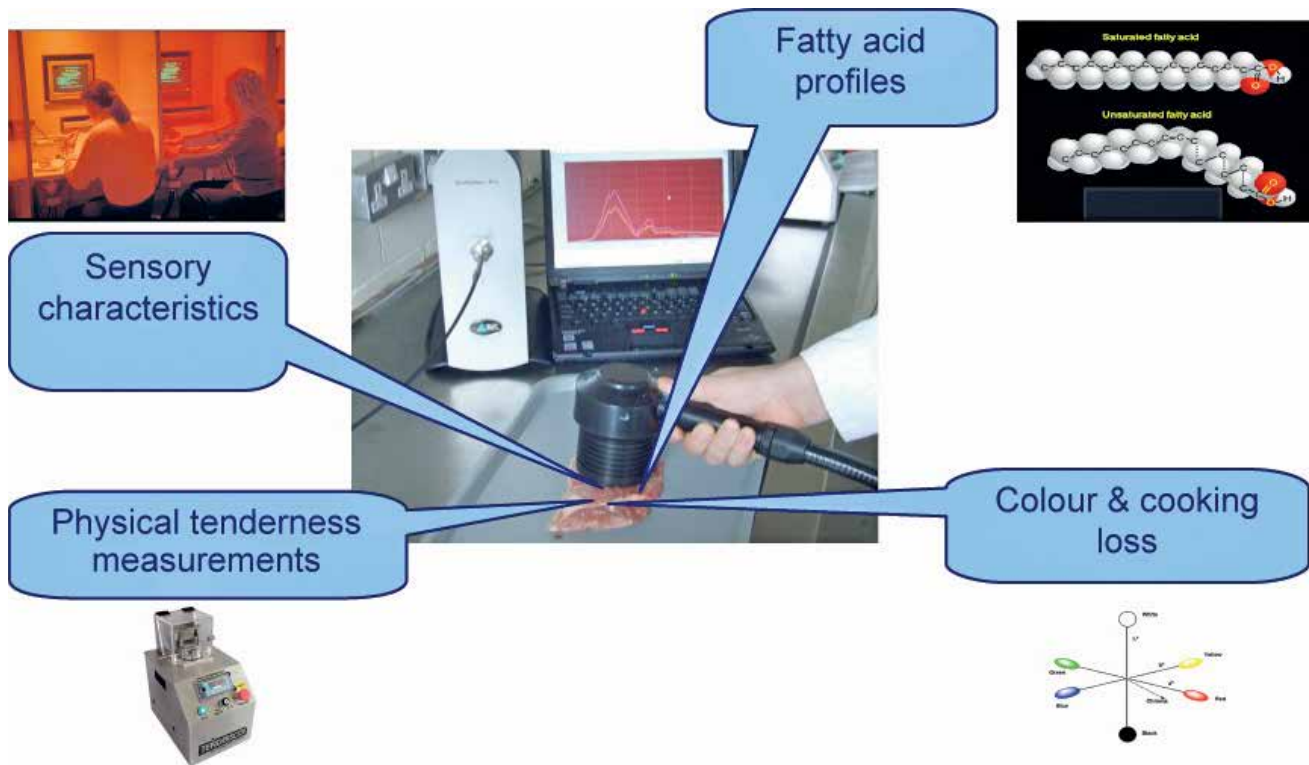
- Computed tomography (CT) is a very accurate and precise measurement technique to determine carcass composition in comparison to physical dissection. CT can be used as the 'gold standard' for the calibration and validation of online systems in the abattoir such as Video Image Analysis (VIA) to determine carcass grading (EUROP conformation and fat classes) as well as lean and fat yields of the entire carcass or its carcass joints.
- VIA of lambs showed 13% higher precision than the use of manual carcass classification scores and can therefore be recommended for carcass grading and determination of lean and fat yields of the entire carcass or carcass joints (Rius-Vilarrasa *et al.*, 2009b).
- Visible Near Infrared spectroscopy (NIR) are able to predict meat eating quality criteria such as sensory characteristics (e.g. tenderness, juiciness, flavour), nutritional quality criteria (e.g. fatty acid profiles), technological quality criteria (e.g. cooking loss) and visible quality criteria (e.g. colour parameters).
- Online measurements obtained by VIA and NIR can be used for a value-based marketing system, genetic improvement programmes and management systems to enhance product quality.

Background

Establishing and evaluating accurate, reliable and objective techniques for measuring or predicting carcass and meat eating quality in farm animals is a key step for improving these traits in the industry. To evaluate the accuracy and reliability of online techniques, accurate reference methods are necessary. For carcass quality the use of physical dissection is the reference method of choice. Because physical dissection is destructive, time-consuming and costly, CT could be a more cost-effective alternative reference method to predict carcass composition without damaging

or depreciating the primal joints. In particular for the calibration and validation of online methods of carcass classification and carcass evaluation, an accurate and reliable reference method is necessary to provide confidence for the use of those online methodologies in the industry. Recently, VIA has been reviewed as an online methodology for carcass classification and evaluation of carcass yield in beef (Craigie *et al.*, 2012). For sheep, the usefulness of VIA for carcass classification and carcass composition has been shown in several studies by Rius-Vilarrasa *et al.*, (2009a,b,c; 2010).

Figure 1. Prediction of sensory characteristics, physical tenderness measurements, colour, cooking loss and fatty acid profiles by NIR. Reference measurements for those traits were obtained by a trained taste panel analysis, slice shear force measurements using Tenderscot, Minolta colorimeter measurements, weight loss after cooking to a tissue centre temperature of 71°C and chemical fatty acid extraction followed by gas chromatography, respectively.



For meat eating quality the sensory assessment of tenderness, juiciness and flavour based on a trained taste panel is the reference method choice.

The estimation of these subjective scores of meat eating quality by objective techniques is challenging and should be predicted as early as possible after slaughtering. Prieto *et al.* (2009b) reviewed NIR

and showed its capability to predict, besides numerous other attributes, eating and nutritional quality of meat and indicated its suitability for online application in the abattoir. Healthy beef is largely related to its fatty acid profiles with increased polyunsaturated fatty acids, in particular Omega-3 fatty acids, being associated with higher human health benefits.

Why work is needed

Carcass quality is of high economic value for production efficiency of beef cattle (fat tissue deposition requires at least 4 times as much feed energy than lean tissue deposition) and the value-based marketing of meat using EUROP carcass classification. Therefore, online systems to predict carcass conformation and fat class such as VIA have been developed to predict carcass grading as well as entire carcass composition in the abattoir. To calibrate and validate those systems, accurate reference methods are necessary for which CT may be the method of choice. To identify the accuracy of CT to predict carcass composition of beef cattle a comparison to physical dissection was carried out. High eating and nutritional qualities of meat are of important to consumers and are therefore of great interest to retailers. As a consequence, the measurement of these attributes of meat quality online in the abattoir is needed to create a feedback system to optimise all factors influencing meat quality from farm to abattoir. NIR shows the potential for providing the prediction of different attributes relating to eating and nutritional quality of meat. In order to achieve this, accurate prediction equations have to be developed based on studies, which have recorded NIR spectra as well as measurements of eating and nutritional quality of the same samples using reference methods such as the trained taste panel for sensory characteristics, slice shear force for objective tenderness measurements, colorimeter for colour parameters and chemical extraction and gas chromatography for fatty acid profiles.

The methods used

For farm animals, CT has been extensively used in sheep and pigs but to much lesser extent in beef. This made it necessary to develop thresholds for the segmentation of fat, muscle and bone in the CT spirals of beef carcass joints and to determine the precision and accuracy of the CT for determination of carcass composition in beef cattle (Navajas *et al.*, 2010a). NIR has been used to predict eating and nutritional quality characteristics of meat under laboratory conditions. However, the use of this technique in an abattoir as early as 48h post mortem is rare and would have direct implementation for the industry. The following results are obtained in studies carried out at SRUC based on CT and NIR using reference methods of physical dissection, trained taste panel analysis, objective tenderness using Volodkevitch shear force and chemical fatty acid analysis carried out at the University of Bristol (Tables 1 to 3).

The results obtained

The accuracies of estimation of carcass composition of beef cattle using CT are very high in the range of R^2 from 0.95 to 0.96 for different tissues (Table. 1). The regression slope is close to one indicating that the developed CT prediction showed no systematic bias in estimation of body composition obtained by physical dissection. Moreover, Navajas *et al.* (2010b) showed that the entire beef carcass composition can be reliably estimated from the tissue weights of a single primal cut assessed by computed tomography.

Table 1. Accuracy and precision of CT to predict carcass composition of beef cattle in comparison to physical dissection (n = 44; Navajas *et al.*, 2010a)

Tissue weights carcass	Regression slope	R^2	RMSE
Fat (kg)	1.002 0.005	0.96	1.28
Muscle (kg)	1.003 ± 0.003	0.96	2.28
Bone (kg)	0.999 ± 0.003	0.95	0.37

Accuracies of NIR to estimate sensory characteristics (tenderness, juiciness and flavour) ranged from R^2 of 0.21 to 0.59, with flavour predicted most accurately (Table. 2). Tenderness, which was measured objectively by shear force techniques were estimated by NIR with substantially higher accuracy than tenderness assessed by a trained taste panel.

In particular, for slice shear force measured at 3 days post mortem, which is close to the NIR scanning at 2 days post mortem, the highest accuracy of prediction was achieved. Colour measurements of meat were highly predictable by NIR.

Table 2. Prediction of meat eating quality characteristics of beef using NIR (n=number of animals, R² = coefficient of determination, SE_{Cal} or _{CV} = standard error of calibration and cross validation, respectively; Prieto *et al.*, 2009a)

Characteristic	N	R ²	SE _{cal}	SE _{cv}
Tenderness	173	0.28	0.56	0.60
Juiciness	174	0.21	0.39	0.41
Flavour	181	0.59	0.34	0.42
Abnormal flavour	172	0.22	0.35	0.37
Overall liking	178	0.25	0.37	0.38
Volodkevitch shear force (N)	172	0.37	11.12	12.70
Slice shear force (3 days pm; N)	176	0.54	46.49	55.76
Slice shear force (14 days pm; N)	176	0.31	26.97	28.49
Cooking loss (%)	130	0.35	2.13	2.35
L* colour	178	0.86	0.88	0.96
a* colour	176	0.86	0.71	0.95
b* colour	171	0.91	0.52	0.69

NIR predicts fatty acids based on the absorption of infrared light by carbon-hydrogen bonds (Table 3). The prediction accuracies were different for Aberdeen Angus than for Limousin. Generally

moderate R² were estimated suggesting the good capability of NIR to predict fatty acid profiles. As well as NIR, CT can predict the fatty acid profiles in beef based on muscle density (Prieto *et al.*, 2010).

Table 3. Prediction of fatty acid profiles of beef using NIR (Prieto *et al.*, 2011)

Fatty acids (FA)	Aberdeen Angus (n = 84)		Limousin (n = 105)	
	R ²	SE _{cv}	R ²	SE _{cv}
Saturated FA	0.40	402	0.68	235
Monounsaturated FA	0.44	452	0.75	240
Polyunsaturated FA	0.16	16	0.64	17
Omega-6 FA	0.73	18	0.45	21
Omega-3 FA	0.43	8.1	0.12	9.0
Intramuscular FA	0.43	1029	0.75	477
C16:0 (palmitic)	0.48	257	0.69	146
C18:3 n-3 (α-linolenic)	0.27	4.4	0.60	3.3
C20:5 n-3 (EPA)	0.26	2.4	0.16	2.7
C22:6 n-3 (DHA)	0.19	0.4	0.36	0.5

The scientific conclusions

The results indicate the high accuracy and precision of CT to determine carcass composition in beef. Therefore, CT can be recommended as a reference method for calibration and validation of online systems such as VIA. The CT may be even more accurate than the physical dissection due to possible variation among butchers involved in

dissection. NIR has the capability to predict numerous eating, visual and nutritional quality attributes of meat in one rapid taken measurement. Due to its online suitability, the method is likely to have a substantial impact on measurements of eating and nutritional quality of meat in the abattoir.

The next steps

The VIA system for beef carcass evaluation should be calibrated and validated using CT to predict the potential advantage of this system in comparison to manual grading of carcasses. To obtain reliable

prediction equations for NIR, large datasets with accurate measurements of important meat quality attributes using reference methods are necessary.

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Estimation of dry-cured ham composition using dielectric time domain reflectometry and its implementation to industry

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Value for industry

- Dielectric time domain reflectometry system can be considered a useful tool to characterize and classify dry-cured ham in industry.
- It permits to determine salt and water contents in dry-cured ham.
- It can be a non-destructive alternative to destructive analytical determinations.
- It could be useful to verify ETG Jamón Serrano specifications concerning salt and water contents.
- It could be used to classify hams according to its salt content and to select those with a reduced salt content which could be labelled accordingly.

Background

Dielectric properties of biological tissues give relevant information of the product's characteristics and composition (Danez *et al.*, 2008). Many authors have shown how the composition of a foodstuff and its influence on the dielectric spectra can be used to predict different quality parameters (Sosa-Morales *et al.*, 2010) or to detect added water in pork and poultry meats (Kent *et al.*, 1996; 2000; 2002). Differences in dielectric spectra between pale, soft and exudative (PSE), dark, firm and dry (DFD) and red, firm and non-exudative meats (RFN) have also been shown (Castro-Giráldez *et al.*, 2010a). Furthermore, it has been demonstrated that the dielectric properties are well suited to determine salt ions from liquid phase or when they are bound to molecules (actin/myosin system) in the raw muscle tissue during the salting treatment (Castro-Giráldez *et al.*, 2010b).

Besides, techniques such as time domain reflectometry (TDR) can measure dielectric properties of samples by the interaction of an electromagnetic step or impulse containing a wide range of frequencies at the same time (Miura *et al.*, 2003). TDR, in combination with multivariate analysis, has been found to be useful to predict different variables associated with loss of quality of stored fish (Kent *et al.*, 2004; 2007).

Why work is needed?

No studies to determine the composition of dry-cured meat products have been carried out using either dielectric spectroscopy or TDR technology. This technology may be of interest for dry-cured ham industry to determine composition online and use this information for a better characterization of the final product.

The methods used

The TDR device RFQ Scan 3.0 (Sequid GmbH, Bremen, Germany) was used to obtain time domain curves from dry-cured ham samples. In this TDR system, a step signal is generated and applied to the material under test via an open-ended coaxial line sensor, which is in close contact with the sample. The TDR applies a step signal with a 100 ps rise time and a repetition frequency of 20 MHz, corresponding to a frequency range from 20 MHz to approximately 5 GHz. The device samples the time domain waveform with a time-base resolution of 10 ps. The obtained TDR curves were related to chemical composition using multivariate statistical analysis in order to develop models for salt and water contents prediction.

The results obtained

Table 1 shows the regression parameters (R^2 , RMSEC and RMSEV) of the predictive models obtained from partial least square regression (PLSR) analysis. The relationship between the measured and the predicted salt content using the developed predictive models is shown in Figure 1. Salt predictions fitted well to the analytical values for all the samples obtained from different areas of the slice with a RMSEV of 0.22%. No effects of dryness or fatness of the sample on the prediction errors were observed (results not shown). Thus, the application of the predictive model for salt content in areas with different intramuscular fat content (in the range studied here, 3.3 to 29.5%) or in a product with different drying levels (in the range studied here, 37.1 to 65.6%) is feasible. Nevertheless, a slight tendency to underestimate salt content in samples with high and low salt content was observed (Figure 1). More experimental work is needed for estimating samples with salt contents higher than 8%.

The relationship between the analytical and predicted water content is presented in Figure 2. Water content was predicted accurately, showing a RMSEV of 1.67%. Neither salt nor water or fat contents of the sample influenced the water content prediction errors.

Figure 1. Relationship between analytical and predicted NaCl content. ----- bisector.

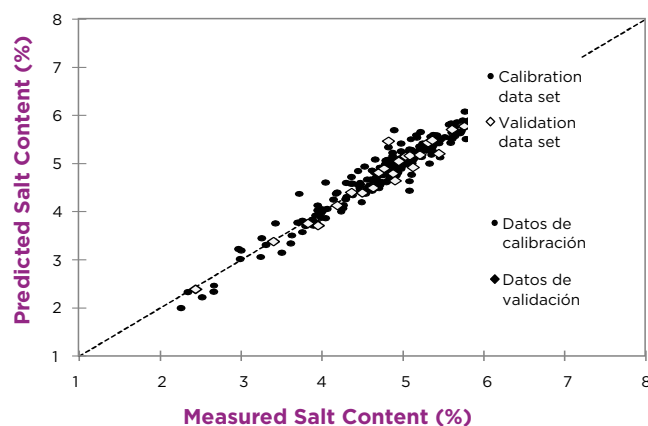


Figure 2. Relationship between analytical and predicted water content. ----- bisector.

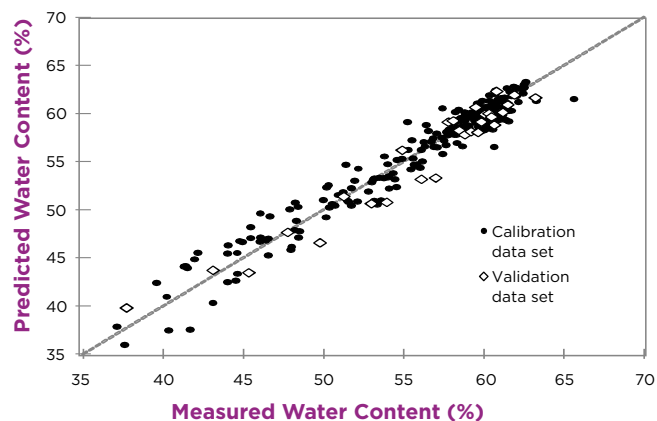


Table 1. R-square and error of the model of prediction (RMSEC) and error of validation (RMSEV) for the prediction models to determine salt and water contents in dry-cured ham.

Models	Calibration				Validation		
	n	R ²	RMSEC	Range	n	RMSEV	Range
Salt (%)	245	0.940	0.217 %	2.3 -7.2 %	27	0.220%	2.5-6.6 %
Water (%)	245	0.948	1.340 %	37.1-65.6 %	27	1.669%	37.7-63.2 %

Although the precision of predictive models was low in comparison to the reference methods, it could be high enough for quality control purposes in industry. A direct application of this technology on dry-cured ham in industry would be the characterization and further classification of sliced dry-cured ham as a function of its salt content. The main drawback of the method is that it gives a local estimation of the studied parameters, the penetration of the microwave being no deeper than 1 mm.

In another study, it has been demonstrated that temperature has an important effect on the sample, mainly for salt prediction, observing the maximum deviation in samples with high water content. Applicability of the TDR method in the meat industry to classify and characterize the product depends also on the homogeneity of the product and on the representativeness of the areas of measurement. For example, estimation of the global salt content of dry-cured ham slices is possible with an error of about 0.37% for salt and 1.89% for water.

The scientific conclusions

Salt content (RMSEV=0.22%) and water content (RMSEV=1.67%) can be accurately determined. Developed predictive models were accurate enough to consider the TDR device as a useful non-destructive tool for characterizing and classifying dry-cured ham online in industry.

The next steps

After this research work, Sequid GmbH developed modules to automatically determine salt and water contents using RFQ-scan device, which are actually commercially available. New utilities using this technology are being studied.

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Use of computed tomography to estimate rib section composition from Holstein bulls and steers

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Value for industry

- Computed tomography (CT) scanning of beef rib sections is an accurate tool to estimate rib total fat, total lean, and intermuscular fat which is useful for the classification of cuts for further processing.
- Marbling can be visualized with CT and its prediction is also possible with a higher error.
- The use of CT for prediction of tissue composition could also be applied to other commercial cuts of interest for the industry.

Introduction

Knowledge of the composition of beef carcasses is important issue for the meat industry as well as for the breeding companies because provides information on carcass value and characteristics for marketing purposes, production optimization, nutritional studies or breeding programs (Navajas *et al.*, 2010).

Composition of beef carcasses or cuts is usually measured by physical dissection because it is the reference method. However, there are other non-destructive, accurate, precise and quick technologies that can also be used to achieve these objectives such as visual image analysis (VIA) and computed tomography (CT) (Craigie *et al.*, 2012; Prieto *et al.*, 2010).

CT is based on X-rays that pass through the body/carcass and are attenuated to different degrees according to the density of the different tissues. The attenuation values are expressed in Hounsfields units (HU) and values are related to the different tissues.

The objective of the work was to evaluate the accuracy of CT to estimate the 9-11th rib section composition in terms of lean and total, subcutaneous, intermuscular and intramuscular fat.

Material and methods

Animals and samples

One hundred and sixty one 9-10-11th rib sections from Holstein bulls and steers were used. Animals were slaughtered at 10, 12 and 14 months of age in a commercial abattoir located in Barcelona, Spain (Marti *et al.*, 2012).

At 24h post mortem, the 9-10-11th rib section was removed, transported to IRTA-CENTA in Monells (Girona) and cut according to Hankins and Howe (1946) for CT scanning and manual dissection (Figure 1). The average weight of the rib sections was 4179.16 ± 736.97 g.

CT-scanning

The rib sections were scanned with a General Electric HiSpeedZx/i CT device. The following images were obtained from each rib section:

- One image in the middle of the section and at the level of the 10th rib using the following acquisition parameters: 10 mm thick, helical 2s, 140 kV, 145mA, 512x512 matrix and reconstruction algorithm STD (Figure 1).

- One image at the level of the 11th rib using the following acquisition parameters: 1 mm-thick, axial 3s, 120 kV, 200 mA, 512x512 matrix and reconstruction algorithm EDGE and DFOV 350mm to visualize marbling (Figure 2) according to Font i Furnols *et al.* (2009) and Brun *et al.* (2011).

In addition, a subsample of 45 rib sections was completely scanned using the same parameters as the first image and obtaining 1 image every 10mm with an average of 27 images per section.

Dissection and intramuscular fat analysis

After scanning, all rib sections were dissected and intermuscular fat, subcutaneous fat, bones and lean were separated and weighed. The average lean content was $2280.52 \pm 391.95g$.

Intramuscular fat was determined by using a FoodScan™ analyzer (FOSS, Denmark) based on near infrared technology. Fat content was on average $2.24 \pm 1.09\%$.

Images treatment and data analysis

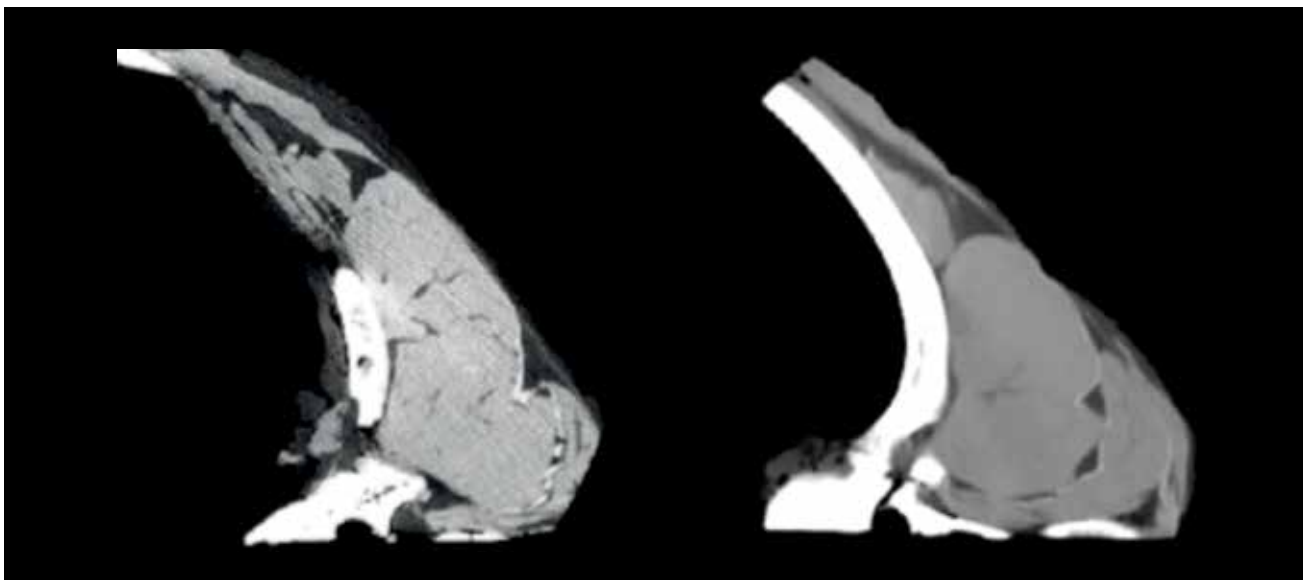
The volume associated with each Hounsfield (HU) value, between -100 and +120 was obtained and used for the prediction.

The volume associated with HU values between 0 and 120 was used for lean determination while those associated with HU values between -100 and 0 was used for fat determination. The percentage of volume between -100 and 0 with respect to the total volume (HU values between -100 and +120) was used for intramuscular fat estimation.

Figure 1. 9-11th rib section (left) and evaluation using CT (right).



Figure 2. Image obtained from the rib section to determine its composition (left) and to visualize marbling (right).



Results and Discussion

Prediction parameters for the 9-11th rib section composition are shown in Table 1. Subcutaneous and intramuscular fat had higher prediction errors as shown by their poorer estimate, due to the lower weight and relative importance of these tissues in the rib section. These results can be expected because the amount of tissue to be predicted is lower and the amount of voxels with partial volume effect is higher, which makes it more difficult to separate the voxels into fat or lean.

In general, the estimation of the 9-10-11th rib composition was better when the whole cut was scanned rather than obtaining only one image per each rib section (see Table 1), mainly because the error of prediction (RMSEP) was lower and the determination coefficient was higher (R^2). The best prediction was for total fat ($R^2 = 0.94$ and RMSEP = 46g) and total lean ($R^2 = 0.97$ and RMSEP = 47g) of the rib section. These results are in line with those found by Navajas *et al.* (2010) and Prieto *et al.* (2010). Intermuscular fat estimation presented R^2 of 0.77 and RMSEP of 56g, and subcutaneous fat had R^2 of 0.53 and RMSEP of 57g which are worse than those found by Prieto *et al.* (2010). This difference in precision of the prediction can be due to differences in acquisition parameters and due to the fact that Prieto *et al.* (2010) took 8 mm-thick images of the cut, while in the present work images were 10 mm-thick.

It is important to find the equilibrium between the precision and the cost of using one image or more than one image or the whole section, because although the use of only one image is less precise it is also less expensive and less time consuming. Thus, the necessities of precision and the budget will determine the methodology to be used.

Estimation of the intramuscular fat had R^2 values of 0.53 and RMSEP of 0.56%. This prediction using the HU distribution of all the rib section was better than using only one image of the *Longissimus thoracis* muscle (results not shown). This is probably due to the fact that the correlation between intramuscular fat and total fat of the cut is 0.60 and because, as reported by Indurain *et al.* (2009), some fatness measurements can improve intramuscular fat estimation. Regarding the prediction of the intramuscular fat using the image obtained at the level of the 11th rib, it is clear that the error is quite high (1.02%) and the determination coefficient quite low (0.18). Thus, this image is good to visualize the marbling but not to predict intramuscular fat content.

Table 1. Prediction parameters of the 9-11th rib section composition.

Prediction of:	R^2	RMSEP
10th rib image		
Total fat (g)	0.87	134.8
Total lean (g)	0.69	219.4
Intermuscular fat (g)	0.66	116.8
Subcutaneous fat (g)	0.52	125.2
Intramuscular fat (%)	0.54	0.74
Between 9th-11th rib images		
Total fat (g)	0.94	45.8
Total lean (g)	0.97	46.8
Intermuscular fat (g)	0.77	55.9
Subcutaneous fat (g)	0.53	57.4
Intramuscular fat (%)	0.53	0.56
11th rib image		
Intramuscular fat (%)	0.18	0.92

The scientific conclusions and next steps

CT scanning of beef rib sections is an accurate tool to estimate total fat, total lean, and intermuscular fat content and the predictions improve when the whole section is scanned rather than a single cross-sectional image. However, the preferred prediction method will depend on the balance between precision and cost. Further work is underway to develop cost effective predictions of rib joint composition using CT scans.

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Prediction of gross composition, salt content and water activity of fresh meat and meat products by mid infrared attenuated total reflectance spectroscopy

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Value for industry

In this study, it was shown that mid infrared spectroscopy has the potential to predict simultaneously gross composition, salt content and water activity of different raw, intermediate and finished meat products.

Introduction

Knowledge of the nutritional value of food products is of great importance to obtain a balanced diet. Increasingly, consumers are paying attention to the quality of food products and making decisions about their food consumption based on the nutrition label. Nutritional claims were designed to encourage manufacturers to produce healthier food products. For example, since a relation between high dietary salt intake and cardiovascular diseases has been demonstrated, consumers' trends are moving to low-sodium food products (Feldman & Schmidt, 1999; Ruusunen & Puolanne, 2005; World Health Organization, 2007). Also the meat industry is trying to reformulate their products by searching for alternatives for sodium chloride (NaCl), such as potassium chloride and sodium lactate, but substitutes must have minimal effects on the water activity (a_w) and the shelf-life of the (Desmond, 2006; Samapundo *et al.*, 2010).

Reference methods to determine gross composition, salt content and water activity are relatively simple but labor intensive and time-consuming to carry out. This ensures that the meat industry is asking for fast and accurate tools to replace the traditional reference methods. Spectroscopic techniques do offer a lot of potential in this regard. Among these light based techniques, mid infrared (MIR) spectroscopy

represents a promising tool. A few studies are available demonstrating the use of MIR spectroscopy to predict gross composition of animal products (Inon *et al.*, 2004; Rodriguez-Saona *et al.*, 2006). Soyeurt *et al.*, (2009) showed that MIR spectroscopy has the potential of predicting different minerals including sodium (Na^+) in cow's milk, while Karoui *et al.*, (2006) used MIR to predict NaCl in cheese. Collell *et al.*, (2011) recently published information relating the use of near infrared (NIR) spectroscopy to determine the a_w value of dry-cured ham.

As the MIR region is based on fundamental vibrations of functional groups of the molecule, MIR spectra are less difficult to interpret and have a better signal/noise ratio compared to NIR spectra which consist of overtones and combination bands (Inon *et al.*, 2004; Carbonaro & Nucara, 2010). Since the intensities of the characteristic absorption bands are proportional to the concentration of the components, MIR can also be used for quantitative analysis (Vlachos *et al.*, 2006).

The aim of this study was to evaluate the use of MIR spectroscopy to determine the gross composition, salt content and water activity of raw, intermediate and finished meat products, especially to implement smooth input and output controls in meat companies.

Materials and methods

A. Meat samples

Both fresh and frozen meat samples were supplied by the companies participating in the project, which are active in the Belgian meat processing industry. Different types of heterogeneous meat products (fresh meat: beef, poultry, pork; fat tissue: lard, backfat; processed meat: fermented sausage, cooked ham, dried ham, pâté) were collected between May 2008 and November 2011. The supplied products were chosen in this way to obtain a wide variety in gross composition, salt content and water activity.

B. Wet chemical analysis

Two heterogeneous datasets were built up i.e. 1) to calculate the prediction equations and 2) to perform an external validation of the prediction equations. Regarding the calibration dataset 96, 100 and 137 meat samples were analyzed for gross composition, salt content and water activity respectively; for the external validation dataset respectively 78, 71 and 94 samples were analyzed. The gross composition was determined by the reference methods, i.e. dry matter content (ISO 1442 - 1973), crude fat content (ISO 1444 - 1973) and crude protein content (ISO 937 - 1978). Reference methods for the determination of Na⁺ and NaCl content were inductive coupled plasma (ICP) and indirect measurement by titration of chloride ions (Method 976.18, AOAC - 1995) respectively. The a^w value was measured at 25°C using the chilled mirror dewpoint technique (AquaLab, Decagon Devices).

C. Collecting MIR spectra in conjunction with sample preparation

The MIR spectra of the homogenized, intact samples were collected between 4000 and 600 cm⁻¹ at a resolution of 4 cm⁻¹ on a Fourier Transform spectrometer (Nicolet X700, ThermoFisher Scientific) equipped with an attenuated total reflectance (ATR) sampling accessory. The ATR sampling technique is based on multiple internal reflectance, and uses a zinc selenide (ZnSe) crystal which enables samples to be examined directly in the solid state without further preparation. Instrument control and spectral collection were performed using OMNIC software (version 6, ThermoFisher Scientific).

In this study, much attention has been paid to preparation of the meat samples. On several model products (pork and backfat) the sample preparation was investigated. After standardizing the different steps in the sample preparation, i.e. completely homogenizing the sample, pressing samples with a standardized weight of 1 gram and standardized temperature of 15°C on the crystal, reproducible spectra were obtained. For each meat sample, 5 spectra were recorded of which an average spectrum was calculated and used for further analysis. The ZnSe crystal was cleaned between measurements with hexane.

D. Statistical analysis

Based on the reference and spectral data, calibration models were calculated using the Partial Least Square (PLS) regression technique and validated using 2 different methods: an internal validation using a leave one out cross-validation and an external validation using meat samples which were not used to build the prediction equations. The accuracy of each calibration was evaluated based on the calibration (RC), cross-validation (RCV) and prediction (RP) correlation coefficient. A correlation coefficient between 0.91 and 0.95 reveals good prediction; calibration models having a correlation coefficient above 0.95 are considered to be excellent (Fagan *et al.*, 2007). Also the errors on calibration (RMSEC), cross-validation (RMSECV) and prediction (RMSEP) were calculated to test the quality of the prediction equations. Sinnavee *et al.*, (1994) considered a PLS model as acceptable with a ratio standard deviation of the reference data on RMSECV or RMSEP, called the ratio of prediction to deviation (RPD), larger than 2. A RPD ratio less than 1.5 indicates that the calibration model cannot be used for further prediction.

Various data pretreatments, using the Unscrambler software, and spectral regions were compared in order to find the most robust calibration models. Mathematical preprocessing techniques such as vector normalization (VN), multiplicative scatter (MSC) correction, standard normal variate (SNV) correction, derivatives of the original spectrum or a combination of these techniques were used to enhance the prediction ability of the prediction equations.

Results and discussion

Statistical parameters of both calibration and external validation dataset are given in Table 1. The selected spectral regions, spectral pretreatment, number of PLS factors and the results for calibration and cross-validation are shown in Table 2. The MIR calibration models, tested by cross-validation, yielded robust prediction equations for gross composition, salt content and a_w with RC and RCV of minimum 0.96 and 0.93 respectively. The RMSECV varied between 2.75 and 9.04 g/100 g for gross composition; the RMSECV was 0.28 and 0.58 g/100g for Na⁺ and NaCl respectively, and 0.01 for a_w .

Good calibration models were obtained based on the RPD value also, which varied between 2.81 and 4.90. A relatively good distribution of the meat samples along the calibration equations was observed. The results for the external validation are reported in Table 3. After validating the calibration models with meat samples which were not used to build the prediction equations, the RP varied

between 0.59 and 0.95. The RMSEP varied between 4.33 and 11.37 g/100 g for gross composition; the RMSEP was 0.39 and 1.01 g/100 g for Na⁺ and NaCl respectively, and 0.03 for a_w . Concerning the external validation dataset, the RPD value was only larger than 2 for water and crude fat content. The prediction was still not accurate enough for crude protein, salt content and a_w due to the high RMSEP.

Due to the lack of samples in specific concentration intervals, relatively poor distributions of meat samples along the prediction equations were observed, especially for crude fat, salt content and a_w . For crude fat content, there is a lack of samples in the range between 40 and 70 g/100 g. Expanding the dataset using artificial mixed samples could possibly improve the model. For Na⁺ and NaCl, a small number of samples at the higher salt concentration was analyzed while for a_w less samples at the lower a_w value were examined.

Table 1. Statistical parameters¹ of both calibration and external validation dataset

Parameter	Calibration Dataset					External validation dataset				
	N	Mean	SD	Min	Max	N	Mean	SD	Min	Max
Water (g/100g)	96	56.54	23.25	7.51	77.46	78	54.68	22.94	5.44	75.56
Crude protein (g/100g)	96	18.11	7.99	1.45	28.86	78	16.53	7.72	1.49	25.64
Crude fat (g/100g)	96	25.26	31.20	0.67	92.34	78	24.51	28.21	0.86	91.28
Na ⁺ (g/100g)	100	1.47	0.81	0.47	3.12	71	1.09	0.45	0.32	2.67
NaCl (g/100g)	100	3.19	1.73	0.79	6.78	71	2.67	1.40	0.93	7.19
a_w	137	0.942	0.049	0.810	0.997	94	0.956	0.039	0.832	0.998

¹ SD = standard deviation; Min = minimum; Max = maximum.

Table 2. Calibration and cross-validation results¹

Parameter	Spectral region (cm ⁻¹)	Pretreatment	PLS Factors	R _c	RMSEC	R _{cv}	RMSECV	RPD
Water (g/100g)	2950 - 900	SNV	12	0.99	4.26	0.96	6.06	3.84
Crude protein (g/100g)	4000 - 600	SNV	17	0.98	1.74	0.94	2.75	2.81
Crude fat (g/100g)	3030 - 2800; 1720 - 620	1 st derivative	14	0.99	5.68	0.96	9.04	3.45
Na ⁺ (g/100g)	4000 - 600	SNV	10	0.96	0.22	0.93	0.28	2.89
NaCl (g/100g)	4000 - 600	MSC	12	0.97	0.42	0.94	0.58	2.98
a_w	3400 - 700	SNV	8	0.97	0.01	0.96	0.01	4.90

¹ R_c = calibration correlation coefficient; RMSEC = root mean square error on calibration; R_{cv} = cross-validation correlation coefficient; RMSECV = root mean square error on cross-validation; RPD = ratio SD on RMSECV; SNV = standard normal variate; MSC = multiplicative scatter correction.

Table 3. External validation results¹

Parameter	R _p	RMSEP	RPD
Water (g/100g)	0.95	8.19	2.80
Crude protein (g/100g)	0.88	4.33	1.78
Crude fat (g/100g)	0.93	11.37	2.48
Na ⁺ (g/100g)	0.59	0.39	1.15
NaCl (g/100g)	0.71	1.01	1.39
a _w	0.69	0.03	1.30

¹R_p = prediction correlation coefficient; RMSEP = root mean square error on prediction; RPD = ratio SD on RMSEP.

The scientific conclusion

Accurate calibration models for gross composition, salt content and a^w were obtained (R_c min 0.96; R_{cv} min 0.93; RPD min 2.81) but after external validation it was shown that the prediction was still not accurate enough for crude protein, salt content and a_w. MIR spectroscopy in conjunction with PLS regression has the potential to predict gross composition, salt content and a_w but optimization of the external validation is necessary, probably due to the heterogeneity of the tested samples.

The next steps

Due to the heterogeneity of the tested products and the lack of samples in some concentration intervals, optimization of the external validation by expanding the dataset is necessary.

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The use of NIR for the prediction of meat quality and fatty acid profile aimed at the genetic improvement of beef cattle

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Value for industry

The beef industry (e.g., Breeders organisations, Herd Books, A.I. centres, PDO - PGI Consortia, Abattoirs, Beef retailers, etc.) can use Near InfraRed Spectroscopy (NIR) as tool for the genetic improvement of beef cattle and for monitoring meat quality over the meat supply chain.

- Beef quality and chemical composition traits, measured by reference methods, exhibits exploitable genetic variation;
- NIR can predict rapidly, cheaply and with acceptable accuracy beef chemical composition (including also fatty acids profile) and some physical traits (e.g., colour traits and drip loss);
- Such predictions shows genetic correlations with corresponding reference measures higher than phenotypic ones; iv) the predictions are more accurate on intact muscles cross sections than on minced samples;
- The use of extended spectra, from visible to mid-infrared, allows for better prediction of beef quality physical traits; vi) there is some perspective to use the spectra collected at abattoir at line on exposed muscles of whole carcasses to estimate beef quality of inner muscles after aging.

Background

Beef quality traits are very important for the whole beef production chain but they are not considered in the selection indices of beef breeds despite exhibiting genetic variation (Boukha *et al.*, 2011). The problem is mainly related to difficulty to collect phenotypes at the individual level. Basically, large-scale recording of individual beef quality traits is critical because the available techniques are time-consuming, and as yet, no high-throughput automated measuring device has been developed.

Why work is needed

Objectives of the research activity at Padova University were/are to test the possibility of obtaining rapid and cheap predictions of meat quality, possibly at the abattoir and at line or off line, for monitoring the beef production chain and implement data collection valuable for the genetic improvement of beef breeds.

The methods used

Five trials have been carried out using samples from the carcasses of the following animals:

- 1,150 Piemontese young bulls (one minced sample from *M. longissimus thoracis* of each carcass used for NIR spectrum collection and reference analyses);
- 1,230 Piemontese young bulls (one minced and one intact subsample from *M. longissimus thoracis* of each carcass used for NIR spectrum acquisition and reference analyses, respectively);
- 21 Chianina, Marchigiana and Romagnola young bulls (15 samples of 15 different muscles of each carcass divided into one minced and one intact subsamples: 672 subsamples in total; NIR spectra were collected on all subsamples, reference analyses were done on intact subsamples);
- 149 Charolais, Limousin and Irish crossbred young bulls (NIR spectra collected on whole carcasses after slaughter and beef quality measured using reference methods on one aged *M. longissimus thoracis* sample per carcass);
- 81 young bulls and heifers obtained from Belgian Blue sire mated to dairy cows (NIR spectra collected on whole carcasses after slaughter and beef quality measured using reference methods on one aged *M. longissimus thoracis* sample per carcass);

All animals were raised in commercial herds in Italy with the only exception of those of the trial 5 reared in the Experimental Farm of the University of Padova.

The physical meat quality traits (i.e., colour traits, drip losses, cooking losses, shear force of cooked meat) were analysed on all samples collected on trials 2, 3, 4, and 5 according with reference methods described by Boukha *et al.* (2011). The intramuscular fat and fatty acid profile have been analysed on 148 meat samples of the trial 1, according to reference method described in Cecchinato *et al.* (2012).

The infrared spectrometers used for the trials were: a) a Foss NIRSystems 5000 (Foss Electric A/S, Hillerød, Denmark) for trial 1 and 2; b) Foodscan (Foss Electric A/S, Hillerød, Denmark) for trial 3; and c) LabSpec2500 (Qualityspec Pro, ASD Inc., Boulder, CO) for trials 3, 4 and 5. The spectrometers work on different wavelength interval, as outlined in Figure 1, as the first instrument cover a range in the NIR - MIR waves, the second a narrow range of NIR waves and the third a wide range in the visible - NIR region.

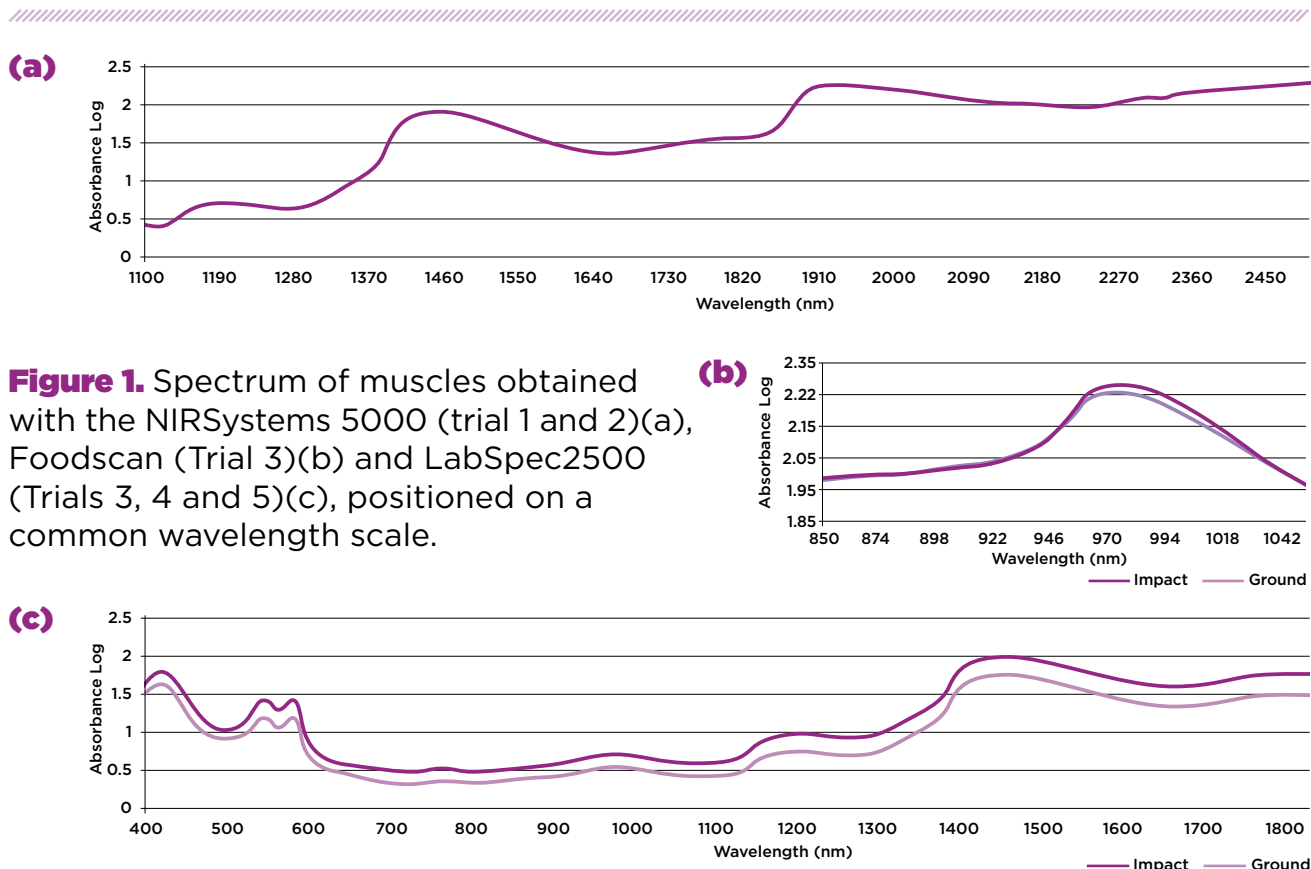


Figure 1. Spectrum of muscles obtained with the NIRSystems 5000 (trial 1 and 2)(a), Foodscan (Trial 3)(b) and LabSpec2500 (Trials 3, 4 and 5)(c), positioned on a common wavelength scale.

To optimize the accuracy of the calibration, deleting of anomalous spectra in the calibration dataset, different combinations of scattering corrections, and several derivative mathematical treatments to reduce the noise effects were applied. Calibration equations were developed from reference data of all investigated meat quality traits using partial least

square regressions. Predictive ability of the models was assessed by coefficient of determination of cross validation (R^2_{cv}) and root mean square error of cross-validation. Prediction models from spectral data were obtained by using the Unscrambler software (version 9.6; Camo A/S, Oslo, Norway).

The results obtained

Prediction of fatty acid profile of meat: NIR calibrations obtained on 148 samples were satisfactory ($R^2 > 0.60$) for 6/8 of the major FA, 6/19 of the minor FA, and for SFA, MUFA, and PUFA (Table 1) and were used to predict FA content of all 1,150 Piemontese young bulls through their NIR spectra.

Heritability of fatty acid profile of meat: Estimates of h^2 for FA predicted by NIR were low to moderate (0.07 to 0.21). NIR is a useful technique (cheap and rapid) to predict the meat content of several FA and FA categories for predicting breeding values of animals (Cecchinato *et al.*, 2012).

Prediction of physical properties of meat: NIR calibrations obtained using spectra from minced samples of *M. longissimus thoracis* were satisfactory for L^* ($R^2 = 0.64$), a^* ($R^2 = 0.68$), hue angle ($R^2 = 0.81$), and saturation index ($R^2 = 0.59$), but not for b^* , DL, CL, and SF (Table 2).

Heritability of physical properties of meat: The loss of phenotypic variability varied from 7% for H index to 85%, being a function of the calibration R^2 . The loss of genetic variability was sometimes lower than phenotypic one and this explains why these traits (b^* , S, and CL) yielded heritability estimates for meat quality predicted by NIR greater than the corresponding values of measured traits (Table 2). The genetic correlation between measured and predicted traits was very high and positive for colour indexes, high for drip loss, and negligible for cooking loss and shear force. These results indicated the possibility of using NIR prediction of colour traits and drip loss for genetic improvement of beef cattle (Cecchinato *et al.*, 2011).

Table 1. Coefficient of determination of cross-validation for FA analysed by GC and heritability estimates for FA predicted by NIR in trial 1.

	R^2_{cv}	h^2
IMF	0.82	0.18
Σ SFA	0.79	0.15
Σ MUFA	0.80	0.21
Σ PUFA	0.61	0.19
Major FA:		
C14:0	0.78	0.19
C16:0	0.83	0.17
C16:1	0.82	0.12
C18:0	0.71	0.21
C18:1n-9	0.80	0.16
C18:1n-11	0.70	0.09
C18:2n-6	0.39	-
C20:4n-6	0.01	-
Minor FA:		
C10:0	0.61	0.16
C12:0	0.63	0.11
C17:0	0.69	0.21
C17:1	0.73	0.20
C18:2 CLA	0.62	0.15
C20:2	0.76	0.21

¹:Other minor FA had $R^2_{cv} < 0.60$

Table 2. Cross validation (trial 2) between meat quality traits measured with reference methods and predicted by NIR, loss of phenotypic and genetic variability of predicted respect to measured traits, heritability of measured and predicted traits and genetic correlation between them.

	R^2_{cv}	$\Delta\sigma_p$ %	$\Delta\sigma_g$ %	h^2_{LAB}	h^2_{NIR}	$R_{G:LAB-NIR}$
L*	0.64	-16	-25	31	26	+85
a*	0.68	-27	-20	32	36	+98
b*	0.44	-42	-6	13	29	+93
H	0.81	-7	-9	63	62	+99
S	0.59	-26	-3	15	23	+95
Drip loss (%)	0.17	-62	-74	24	14	+72
Cooking loss (%)	0.04	-85	-72	5	17	-4
Shear force (N)	0.21	-61	-60	10	10	-10

Calibration of physical properties of intact and minced meat using NIT and NIR: The NIR transmittance lab. instrument, using narrow spectra and set up for chemical analyses, gave unreliable predictions for physical meat quality traits when spectra were collected on minced muscles (Table 3). Cross validation improved moving to spectra from intact muscles, improved further using a NIR absorbance portable instrument on minced meat and again on intact muscles (De Marchi *et al.*, 2013).

Prediction of physical properties of intact aged muscles using NIR spectra collected on intact carcasses after slaughtering: The preliminary unpublished results (De Marchi, 2012, personal communication) gave low to moderate cross validation according to breed of animals and especially to the interval between slaughtering and spectra collection.

Table 3: Table 3: Cross validation between meat quality traits measured using reference methods and predicted by NIT or NIR spectra collected on minced or intact muscles (15 muscles per carcass) (Trial 3).

R^2_{cv}	NIT		NIR	
	minced	intact	minced	intact
pH	0.29	0.31	0.42	0.62
L*	0.34	0.45	0.55	0.70
a*	0.34	0.37	0.52	0.73
b*	0.14	0.20	0.41	0.60
Drip loss (%)	0.01	0.15	0.12	0.15
Cooking loss (%)	0.12	0.31	0.12	0.38
Shear force (N)	0.01	0.15	0.13	0.34

L* = lightness
a* = redness
b* = yellowness
H = hue angle
S = saturation index

The scientific conclusions

The NIR techniques have some potential for a cheap and rapid prediction of meat quality traits to be used for genetic improvement of beef breeds also on intact muscles and carcasses.

The next steps

The next steps will be: i) to extend the prediction on intact muscles to chemical properties, including fatty acid profile, conjugated linoleic acid (CLA) isomers, and cholesterol content; ii) to compare lab and portable spectrometers of different spectrum extension; iii) to define the operational condition allowing an improvement of calibrations equations of spectra collected on whole carcasses for predicting meat quality of aged beef; iv) to study a breeding scheme, specifically for beef breeds, aimed at incorporating meat quality predictions collected at abattoir.

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Computed Tomography image analysis methods used in Hungary

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Value for the industry

According to our experiences in agricultural CT image processing, especially segmentation, it can be concluded that:

- Extracting simple statistical descriptors, characterizing the voxel neighbourhoods by the descriptor vectors and building multivariate statistical models for segmentation can enhance the accuracy of processing.
- By enforcing connectivity, hidden Markov random fields based segmentation methods provide accurate results.
- Morphological operators can efficiently remove noise from label images;
- There is need to develop segmentation methods that provide subpixel-scale accuracy, preferably using and fitting analytic models to object boundaries, instead of the currently applied volumetric approaches.
- There is also a need to develop segmentation methods that do not depend on the exact Hounsfield-units of the images, instead, the relative differences of voxel values are used to determine the object boundaries.

Introduction

Over the last decades, computed tomography (CT) has become a general tool for research in agriculture. Although several algorithmic solutions have been developed for the processing of medical images, agriculture requires new approaches for several reasons. Unlike in medical issues, where semi-automatic methods are used by experts to analyze the CT image of one patient at a time, agriculture needs the accurate processing of many images, to enable the drawing of statistically relevant conclusions. Besides the amount of data to process, the lack of texture differentiates the processing of CT images and medical or simple photographs. The aim of processing is also different in medicine and agriculture. In medical applications, usually the

detection of objects or the proper segmentation of manually selected regions is required, while in agriculture the estimation of volumes and surfaces of various tissues is desired. Since a wide variety of segmentation techniques is published, the choice and adaptation of the proper method for CT images is not obvious. In the rest of the paper the segmentation techniques that have been applied in various CT segmentation problems will be discussed and to provide a better impression of the results, the methods used will be exemplified via the segmentation of a CT image of a hen egg. Figure 1.a shows the CT image and figure 1.b shows the ground truth manual segmentation. The technical details of the developments are described later in this article.

Overview of the Applied and Adopted Segmentation Techniques

In general, machine learning techniques are used to build statistical models for segmentation. In these approaches the result of segmentation is a volumetric label image, that is, specific class (label) is assigned to each voxel and the volume of a tissue or surface of an object can be determined by the sum of voxel volumes or surfaces having the label of the object of interest. To build proper statistical models, a manually segmented database is required.

Preprocessing. In many image processing applications, the first step is some sort of contrast enhancement and noise removal to improve the accuracy of further processing steps. In general, there appears to be little effect of preprocessing methods on segmentation. The application of linear intensity transformations does not improve the separability of tissue classes. To apply global or local non-linear transforms which enhance the separability of the tissues, the transforms should be able to distinguish the tissues which can hardly be done as preprocessing, since exactly this is the aim of the further processing steps.

Bayesian segmentation. Bayesian segmentation methods are based on the estimation of conditional distributions of voxel values for class labels. Then, the class of further voxels can be determined by the Bayesian-rule:

$$L(v) = \operatorname{argmax}_{c \in \text{Classes}} P(I(v)|c) = \operatorname{argmax}_{c \in \text{Classes}} \frac{P(c|I(v))P(I(v))}{P(c)},$$

where v refers to a voxel, $I(v)$ and $L(v)$ denotes the intensity value (Hounsfield-unit) and label of voxel v , respectively. The probabilities of the right hand side can be estimated by histograms or replaced by the probability density function values of fitted Gaussian- or Mixture of Gaussian distributions. The advantage of the approach is the easy implementation and interpretation, while the drawback is the quiet low performance. Figure 1.c shows the Bayesian segmentation of the test image, when the statistical models are trained using 3 manually segmented images.

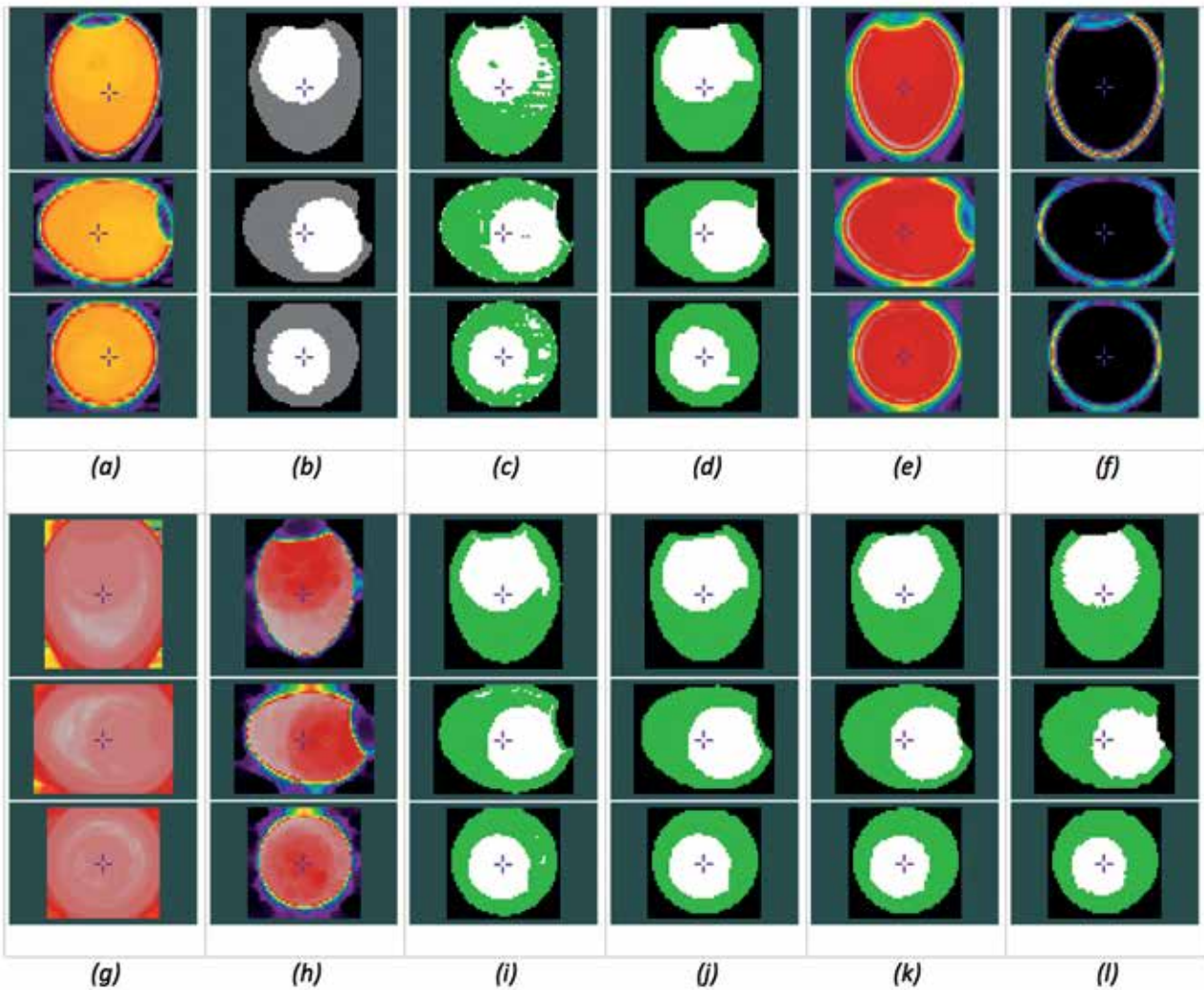
Hidden Markov Random Fields (HMRF) are special probability structures used in a wide range of image processing applications (Zhang *et al.*, 2001). In general and simple form, disregarding the stochastic model behind, the segmentation is turned into an optimization problem: the search space of the optimization contains all the possible labeling of the voxels and the target function is the sum of two terms, particularly, the probability of the given labeling and a penalty term penalizing the differences of neighboring labels. In details, the segmentation is the result of the following optimization:

$$S = \operatorname{argmax}_{L \in \text{Labeling}} \sum_{v \in V} P(I(v)|L(v)) + \beta C(v,L),$$

where V denotes the set of all voxels, β is the penalty parameter and function C gives the number of neighbors of v in L , having different label than v . When β is set to be 0, we get back the simple, Bayesian segmentation. β can be learned from the manually segmented database and the optimization problem can be solved by some kind of stochastic optimization method, like simulated annealing (Kirkpatrick *et al.*, 1983) In general, the results are better than in the case of Bayesian segmentation, the resulting label image contains connected components without much salt-and-pepper like noise, however, depending on the size of the image, the solution of the optimization problem can take a long time and the method is quiet sensitive for the selection of β . Figure 1.d shows the HMRF-segmentation of the test image, using the same statistical model used in Bayesian-segmentation.

Features. In several image processing solutions, the voxel neighborhoods are represented by so-called feature vectors to reduce the size of the neighborhood. In general, *feature* can be any intensity transform, which maps a new intensity to the existing ones, preferably non-linearly. The advantage of features is that they represent the voxel neighborhood, thus, can contain more information than the intensity of a voxel itself. However, one have to find the proper features for a given problem, there are no best features that fit all the segmentation problems. We have tried the simplest statistical descriptors, like the mean, variance, signal-to-noise ratio, min, max values and invariant Hu moments [3], computed in a small radius sphere around the voxels.

Figure 1. Axial, coronal and sagittal slices of the CT image of an egg (a), its manual (b), Bayes- (c), and HMRF based segmentation (d). Images of features, computed in a spherical neighborhood of radius 3, voxel-wise: mean (e), variance (f), signal-to-noise ratio (g), minimum value (h). Segmentation with multivariate Bayesian- (i) and HMRF-model (j), using features. Morphological postprocessing of univariate Bayesian-segmentation (k). The result of incremental Otsu thresholding followed by postprocessing (l).



From the feature vectors multivariate Bayesian and HMRF models were built and we have found that the use of features can enhance both the Bayesian and HMRF segmentation models; however, some features can have large computing demands. Figure 1.e, 1.f, 1.g, 1.h show some feature images and Figure 1.i and 1.j show the Bayesian- and HMRF-segmentation, using feature vectors.

Postprocessing. After the segmentation is performed, the label image can still be noisy. To handle this issue, we have successfully applied the tools of mathematical morphology (Najman and Hugues, 2010). The complex morphological

operations, like opening and closing, bright- and dark-reconstruction can remove salt-and-pepper like noise; however, care must be taken, since these morphological operations do not use the original image content, at all. Thus, important features can be removed from the images, if the size and shape of the structuring element is not properly chosen. In the case of the egg, the shape of the yolk and albumen is both sphere-like, convexly curved, therefore the use of sphere structuring elements of small radius (3) is reasonable. See Figures 1.k for the result of bright-reconstruction, dark-reconstruction and closing applied to the intensity based Bayesian-segmentation (Figure 1.c).

Thresholding. Adaptive thresholding methods are among the simplest techniques to divide a distribution to two parts. In the literature, plenty of adaptive thresholding methods are available (Sezgin and Sankur, 2004), local and global techniques, histogram and image statistics based approaches, etc. Although these methods are usually fast, one limitation of thresholding is that only binary separation can be performed. Another drawback is that most of the adaptive thresholding methods works only if the foreground and background have approximately the same number of voxels. On the other hand, one advantage of thresholding is that many of the methods do not have parameters, and in these cases, no manually segmented databases are required. For the parametric thresholding methods the best parameterizations can be selected using

the manually annotated database. In the case of the eggs, one of the easiest and most popular method, called Otsu-thresholding was applied incrementally: first, the empty background and the egg, then, the albumen and the yolk are separated. See Figure 1. shows the postprocessed results.

Technical details

From the technical point of view, all the methods are implemented in *C/C++* as command line applications and *bash* shell scripts are used for fast prototyping in *Linux* operating systems. The segmentation techniques are implemented in the open source library called *OpenIP* (Kovacs *et al.*, 2010) available at code.google.com/p/openip.

The scientific conclusions and next steps

Application of CT in agriculture needs the accurate processing of many images to enable the drawing of statistically relevant conclusions. Following acquisition, CT images must be processed in order to extract meaningful information; this commonly involves a series of image segmentation steps. A wide variety of image segmentation techniques have been published, but the choice and adaptation of

the proper method for CT images of animals is not obvious. Several image segmentation techniques (including preprocessing, Bayesian segmentation, feature selection, post processing and thresholding) can be implemented. Future work will continue to evaluate how these various techniques can be used together to streamline the extraction of meaningful information from CT scans of farm animals.

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Norsvin imaging methods

J. Kongsro

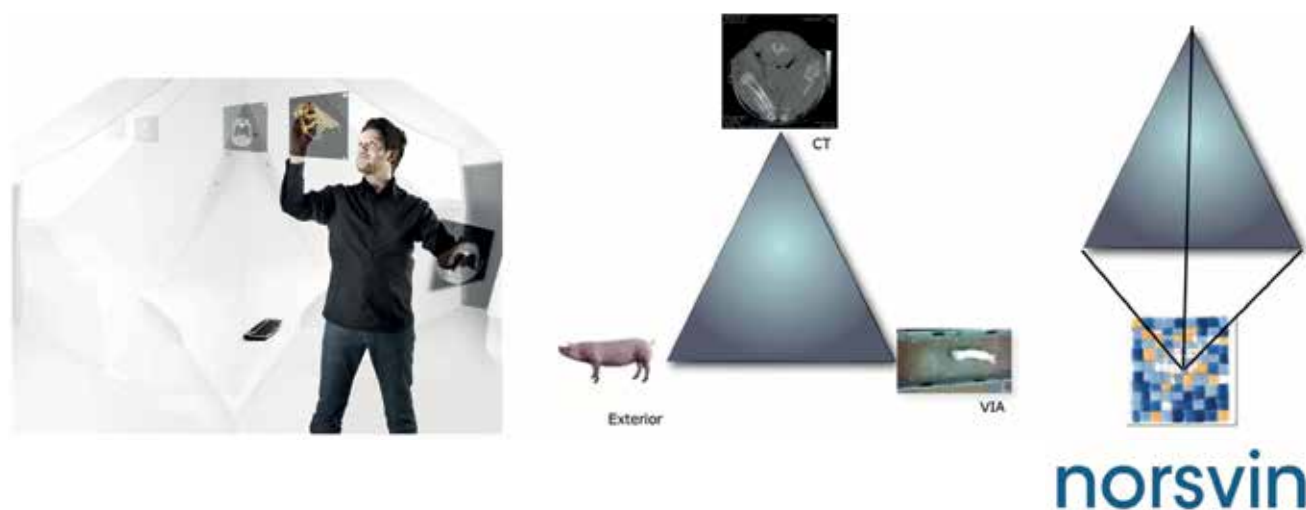
Norsvin, P.O. Box 504, N-2304 Hamar, Norway

Value for industry

Our goal is to identify non-destructive, fast, reliable, affordable *in vivo* methods for sampling phenotypes in pig breeding on body composition, meat quality and leg weakness. Imaging methods will provide important information about the farmed animal, from a clinical perspective to production traits.

- Increase the precision of *in vivo* measurements.
- Provide applications; from automatic solutions to user-friendly diagnostic tools.
- The imaging techniques used in pig breeding for Norsvin includes Computed Tomography (CT), near infrared spectroscopy (NIR), ultrasound and video image analysis (VIA).

Figure. 1. Application of imaging techniques in pig breeding; providing phenotypes as a basis of selection by quantitative genetics or genomic selection.



Background

The Norsvin testing system of terminal boars include state of art technology for measuring feed efficiency, body composition, and meat quality and leg weakness. These traits are considered the most important traits of a terminal boar, and require high quality to phenotypes to get as accurate genotypes as possible. The testing system include Feed Intake Recording Equipment (FIRE®), Computed Tomography for body composition and leg weakness, ultrasound combined with CT and NIT spectroscopy for meat quality and video image analysis for gait scoring and leg weakness. The different technologies require an infrastructure to deal with image analysis and human-computer interactions. Most of the traits is measured automatically (FIRE, body composition and meat quality). However, some require human interaction, like leg weakness scoring using CT.

Osteochondrosis is monitored and recorded manually by using CT, monitoring changes in articular cartilage and bone in fore-and hind limbs. The aim is to develop an automatic system for recording leg weakness based on CT and gait scoring using video image analysis. Outside the test station, in our nucleus farms, ultrasound scanning is used to obtain body composition in the gilts. The gilts are the future mothers of the terminal boars selected for the test station. This requires a fast and efficient system using image analysis which needs to be portable and user friendly. More efficient systems for phenotype collection in our nucleus farms are under development using imaging and vision systems. Weighing of animals, counting of piglet and recording of animal behaviour are tedious tasks which can be done more efficiently. With the advent of genomic selection in the breeding industry, more robust phenotypes are crucial. Consequently, vision and imaging systems can also be applied in commercial farms, where it can assist the farmer obtaining high profits and increase the focus on animal welfare.

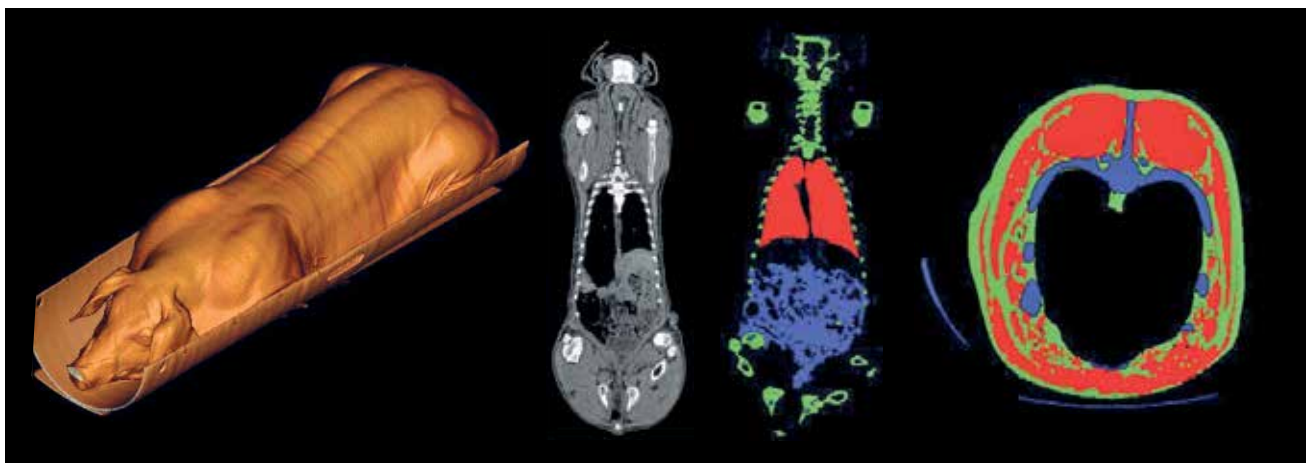
Why work is needed

Engineering and sensor development, including imaging systems, are playing an increasingly important role in automating routine labour activities associated with livestock rearing, selection, breeding and genomics (Deshazer *et al.*, 1988). Imaging or vision systems can provide an addition or replacement of observing or measuring animals. From production traits like piglet weighing and behaviour, to boar selection by the use of CT to estimate body composition, work is needed to develop both hardware and software applications. The hardware needs to be robust and reliable in a farm environment. The software must cover image processing methods, image analysis, pattern recognition and feature extraction. The combination of biological knowledge, hardware and software development, require interdisciplinary knowledge and collaboration. New and innovative developments have to challenge established traditional views within the field of animal science to overcome the challenges of modern breeding methods, size and efficiency of animal production and biosecurity.

The methods used

Computed Tomography was introduced in the field of animal science in the early 1980's by Skjervold *et al.*, (1981). There have been trials conducted both *in vivo* and *post mortem* (carcass and meat products) since then. Norsvin was the first breeding company to apply CT in a large scale testing system for breeding boars, replacing dissection of sibs or half-sibs. The CT provides estimates of body composition (lean meat and yield), meat quality (IMF) and leg weakness (osteochondrosis).

Figure 2. The Norsvin CT application. Estimation of body composition.



Ultrasound has been applied for estimation of back fat and muscle thickness in Norsvin since the 1960's. Today, ultrasound is used in off-test of sows in our nucleus herds, using a B-mode probe for ultrasound scanning. Near-infrared spectroscopy is being used to predict meat and fat quality of meat and fat samples non-selected boars. The results show that both meat and fat quality predicted very accurately using NIR (Gjerlaug-Enger *et al.*, 2011).

Video image analysis is currently under development to study the movement of pigs related to leg weakness. In addition, we are looking into developing a monitoring system for piglet weighing in our nucleus farms.

The results obtained

There has been a small increase in accuracy when replacing dissection with CT. Some results have been published by Gjerlaug-Enger *et al.*, (2012). We have experienced a higher genetic improvement through:

- Sampling carcass traits directly from the live animal.
- Higher reliability (increased heritability on carcass traits).
- Increased number of animals tested.
- New traits can be implemented based on historical data (images stored in archive).

The scientific conclusions

As shown by Gjerlaug-Enger *et al.* (2012), the carcass traits are very heritable measured by CT, achieving a heritability of lean meat percentage of 0.5 to 0.6. Leg weakness has also increased in heritability from 0.1 to 0.3 using CT (Aasmundstad, unpublished data 2012). Meat and fat quality measured using CT is under development, and will be published shortly. By measuring fat and muscle depth by ultrasound more objectively by imaging software, the heritability for fat and muscle depth of sows is also expected to increase. More work is still needed here. Video image analysis is also under development, and data is being gathered for analysis.

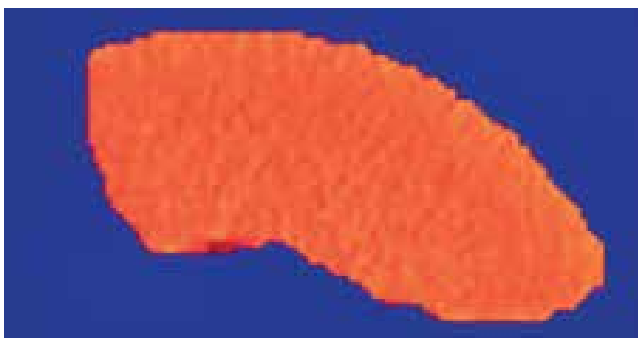
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The next steps

The challenge is to extract relevant information from complex patterns in pig movement. The use of video or vision systems for piglet or pig weighing has been proven accurate (Doeschl-Wilson *et al.*, 2004). The main challenge is to ensure the ID and tracking of animals, to confirm the identity of the animal being measured. Combining vision systems with RFID or other tracing technologies is crucial to setup a robust and reliable system for measuring animals in large scale systems.

Figure 3. Combining NIR with CT and /or ultrasound to obtain in vivo prediction of meat quality.



Semi-automatic 3D segmentation

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Value for industry

Medical imaging systems can be used to reveal the body composition of farm animals, including musculature and distribution of fat deposits. This information is of use in breeding selection and potentially in optimising husbandry and market targeting.

Background and why work is needed

Segmentation, to identify specific tissues, is a key step in converting digital images into usable information. Automatic methods are successful for segmentation in tightly specified circumstances, such as 2D ultrasound and Computed Tomography (CT) images of particular breeds of sheep at specified anatomical locations (Glasbey and Young, 2002) and 3D spiral CT using a single scanning protocol (Glasbey, 2009). However, these methods fail to produce acceptable results if circumstances change, such as images of other breeds/ages of sheep, or from other CT scanners using different protocols, or even images from other species and technologies, such as MRI of pigs! The alternative is manual segmentation, where boundaries are placed on images by human control of a screen cursor. This is especially time consuming and tedious in the case of 3D images, taking several hours of human input to segment a single animal.

Semi-automatic segmentation is a half-way house, where some tasks are performed manually and others automatically by the computer (see, for example, Inglis and Gray, 2001). In 3D segmentation, where a series of slices are to be segmented, each slice is often very similar to the preceding one. Can the computer segment a slice by learning from manual segmentation of the previous slice?

The methods used

The answer is yes, an algorithm has been developed with the following steps:

- a human operator segments image 1 manually;
- the computer segments image 2 automatically, by mimicking the image 1 segmentation (see Fig 1 for an illustration);
- the operator corrects any mistakes in the image 2 segmentation;
- the computer segments image 3 automatically, by mimicking the image 2 segmentation;
- the operator corrects any mistakes in the image 3 segmentation; etc.

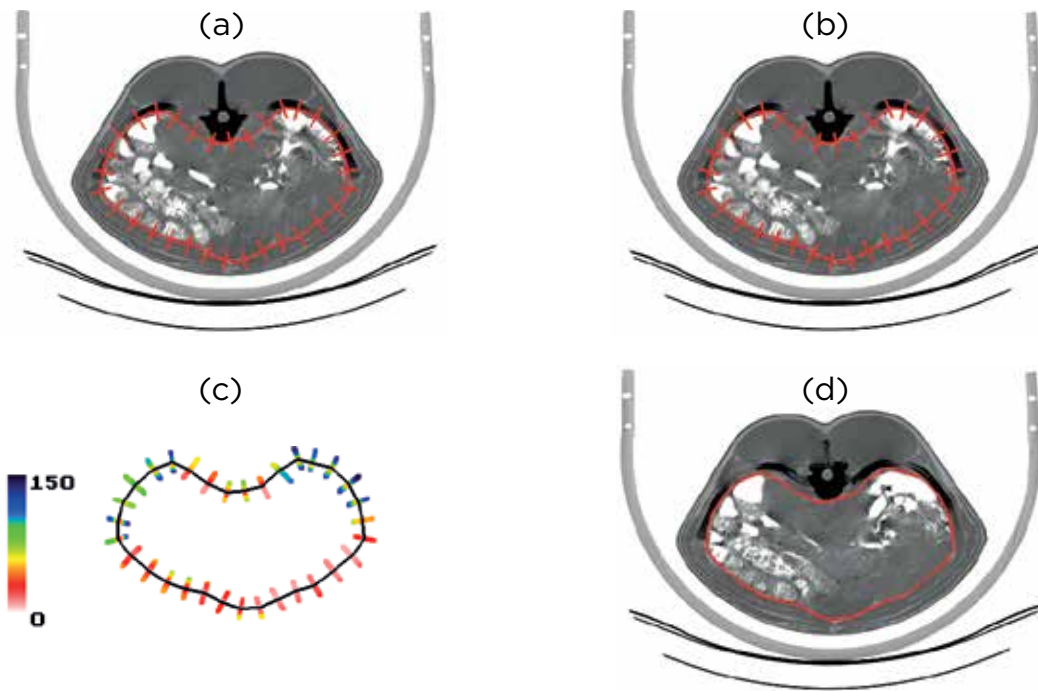


Figure 1. Illustration of how algorithm mimics the segmentation of one image to segment the next one in a 3D image stack, in this case identifying the internal organs in a pig's thorax imaged by CT: (a) image 1 with red lines showing correct boundary + perpendicular transects; (b) the same transects superimposed on image 2; (c) pseudo-coloured score showing agreement between CT values along transects in images 1 and 2 at a range of shifts in alignment, together with smooth black boundary showing minimum cumulated score through all transects found by Dynamic Programming; (d) this boundary superimposed on image 2, showing the algorithm's attempt to mimics image 1 segmentation.

The results obtained

The method is very flexible, as boundaries are allowed to either outline objects or to specify holes within objects, and no prior assumptions are made about patterns of pixel values near boundaries. The computer algorithm perturbs boundaries, and tries to replicate patterns of pixel values, from the previous image.

The scientific conclusions

Experience to date indicates a reduction in human input time from several hours to a few minutes per animal.

The next steps

The basic algorithm is open to many generalisations, such as:

- rules for including/excluding air/bone in segmentation;
- using previous two images and 3D roughness penalty;
- exploiting accumulated database of manual corrections.

Acknowledgements

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New X-ray Imaging Techniques for safe and high quality food

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Value for industry

- New X-ray techniques based on phase contrast and dark field imaging are being developed that will allow objects that have similar absorption profiles, but different microstructures to be distinguished from one another.
- X-ray dark field imaging potentially offers a new technique that can visualize microstructures like muscle and fibres characteristics and hence might be a tool to assess meat tenderness.
- Establishment of a new centre is proposed that aims to promote a synergic alliance between sensor physics, automated image analysis and high capacity computing to encourage researchers and SMEs to interact to develop solutions for industry.

Background

Product quality control is a vital part of the Danish food industries today. High quality products constitute the main of part of Danish exports and making Danish food industry more competitive. Higher quality food requires sensitive and efficient techniques that assess the quality early in the production process using non-destructive modalities. Optical inspection is widely applied but is far from sufficient as it only will probe the surface of the product. In many applications three dimensional techniques, able to analyze the interior of the food product are necessary. For instance, in the meat industry, quality control is laborious and often destructive with respect to the tested sample. Therefore the quality control process is often seen as a costly exercise comprising spot testing and sparse sampling. Techniques are also required to test and

inspect packaging. Traditional X-ray scanning offers, even at relative low photon energies, the penetrating power to monitor the inside of a food product even inside the final packaging, but often does not have the sufficient sensitivity to distinguish subtle details as the measurement contrast arises from small differences in x-ray absorption given by the atomic elements in the sample. We propose to develop new x-ray techniques based on phase contrast and dark field imaging using the recently developed x-ray grating interferometer (Weitkamp et al., 2005; Pfeiffer et al., 2008) that will allow different contrast mechanisms to distinguish objects that have similar absorption, but different microstructure.

Objectives

Production of safe and high quality food products at an industrial scale has been a substantial contribution to Danish export for many years. The production is automated to a large extent to be able to meet the competition from developing suppliers from South America and the Far East.

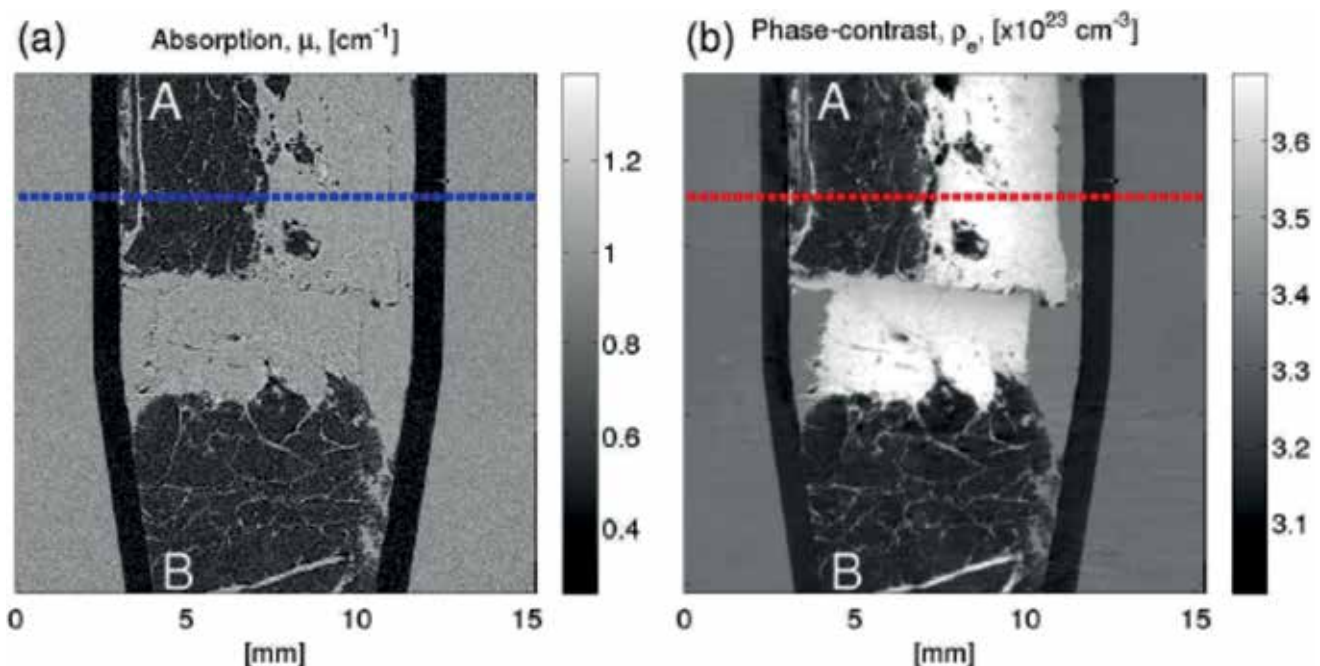
The competitive strength of Danish food production is based on research in most aspects of the entire production chain from farm to fork. This research includes CT scanning for automated assessment of fat and meat composition in pig carcasses, and X-ray screening for foreign material to verify the highest level of food safety. This work has demonstrated the challenges of detection of fatty acids in subcutaneous fat tissue and the detect ability to detect light plastic fragments from broken production tools that might incidentally be present in food products. Preliminary results show an encouraging potential of detection of fatty acid composition, high contrast between cartilage and soft bones, even connective tissue may be discernable in meat products.

It is interesting to investigate the potential of characterizing various quality traits of food products in particular meat quality using the newly invented grating interferometer setup (Weitkamp *et al.*, 2005; Pfeiffer *et al.*, 2008).

These aspects include tenderness and intramuscular fat distribution in bovine meat, subcutaneous fatty acid composition of porcine fat and drip loss of porcine meat. Magnetic Resonance Imaging (MRI) today defines the golden standard for determination of such delicate parameters in food. But as the MRI is sensitive to the proton content of the material many of the interesting meat traits as taste, water holding capacity and tenderness are measured as indirect parameters. With X-ray phase contrast; however, the electron density is directly measured, which can directly be linked to the carbohydrate, fatty acid and protein composition (proteomics) of the tissues (Manohara *et al.*, 2008).

Furthermore, the new techniques will be tested for their ability to detect foreign materials, such as small paper and plastic fragments in minced meat and hence will be tested towards increased food safety. Due to the very high production speed in most food industries, the need for high capacity computing for image construction and evaluation is huge and ever increasing. The implementation of automated detection and assessment algorithms are required even today and with the emerging imaging modality the requirements for fast computing are tripled.

Figure 1. Tomographic slices of porcine fat. (a) Frontal slice through the tomographic reconstruction of the absorption contrast. (b) Frontal slice through the tomographic reconstruction of the phase. The phase gives clearly higher contrast. From Jensen *et al.*, (2010)



Methodology: New X-ray techniques

Today X-ray imaging is based on absorption contrast as it was in the days of Wilhelm Röntgen more than 100 years ago. The contrast between different tissues of the objects arises either due to higher density or heavier elements, both giving higher absorption. However, X-rays are electromagnetic waves and the index of refraction in biological tissues contains an absorption term and a phase term. The refractive index can be written as $n=1-\delta-i\beta$ where δ describes the phase term and β the absorption term. The phase term δ is proportional to the electron density and is about 10^{-6} . δ is a factor 100 larger than the absorption term β , which typically is of the order 10^{-8} . The phase term hence offers potentially much more sensitivity if it can be measured. The phase term is given rise to refraction effects, but typical refraction angles are very small and are normally only detected using partial coherent beams. However, recently a new technique using a grating interferometry technique from optics has been invented by the groups of Prof. F. Pfeiffer in Munich, which makes phase contrast and dark field imaging much more practical. We have participated in collaboration with the group of Prof. Pfeiffer developed the technique and shown its feasibility for medical and food applications (Bech *et al.*, 2009; Jensen *et al.*, 2010, 2011).

In brief, the techniques use a phase grating using the Talbot (Weitkamp *et al.*, 2005) effect to produce an interference pattern. Placing a sample into the beam distorts the interference pattern and the distortion is proportional to the derivative of the phase of the X-ray beam passing through the sample. Taking images at different rotations of the sample a full 3-D reconstructed of the sample can be obtained using standard filtered back Fourier transforms.

The phase grating has a pitch of typically 4 microns. The interference pattern produced by the phase grating has a pitch of 2 microns and cannot be resolved by standard X-ray imaging devices. Instead, it is measured using an absorption grating with about half the pitch of the phase grating stepping the absorption grating across the interference fringes. The total measured X-ray intensity is measuring the absorption, the distortion measures the phase derivative and amplitude of the interference fringes measures the visibility. For samples with a microstructure that give rise to scattering, the visibility will decrease. Hence the technique can be used to measure the scattering from the sample equivalent to dark field imaging known from optical microscopy.

Figure 2. Schematic view of the experimental setup. The G0 grating acts as a slit array, producing parallel line sources. The sample is placed immediately in front of the interferometer, and the detector is placed immediately after. G1 is the phase grating and G2 the absorption grating. From Bech *et al.*, (2009).

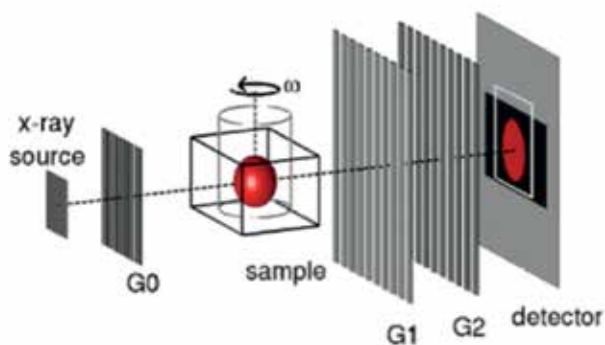
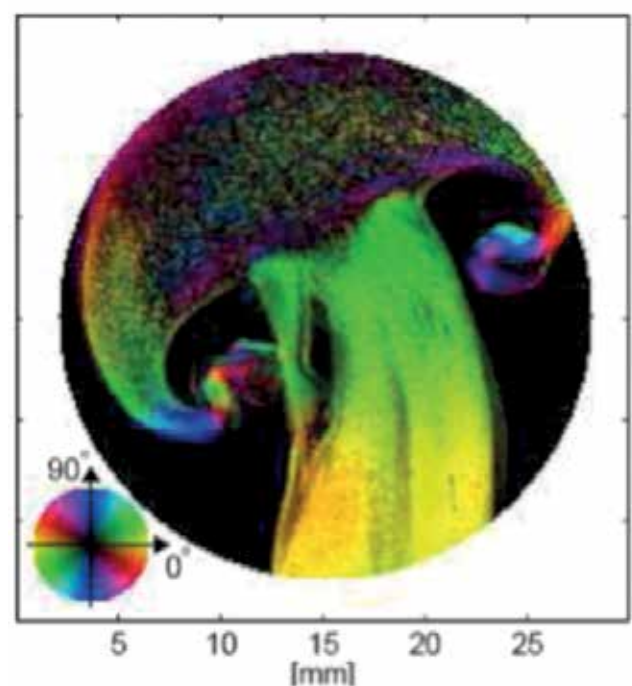


Figure 3. Dark field imaging of the fiber orientation in a mushroom slice. From Jensen *et al.*, (2010).



As the scattering arises from the microstructure of the sample, X-ray dark field imaging potentially offers a new technique that can visualize microstructures like muscle and fibres characteristics and hence might be a tool to assess meat tenderness, see Figure 3. The grating interferometer requires partial coherence of the X-ray wave beam. This can be obtained by using X-ray radiation from large scale international synchrotron radiation facilities like the ESRF in Grenoble. The quality of the X-ray beam in terms of coherence is measured via the visibility of the interferometer and large visibility up to 60% can be measured at synchrotron radiation sources. The limitations of synchrotron radiation are that such facilities only exists few places in Europe and that access is limited. Furthermore due to the high collimation of the beam the field of view is limited to a few centimetres at most. Therefore these facilities will only be used for high specialized research and development.

However, converting a standard laboratory X-ray source into a partial coherent source can be obtained by placing a fine space source grating just in front of the sources (Pfeiffer *et al.*, 2006), see Figure 2. Although presently optimised for small animals, acquiring a setup for use in the Danish food industry will give research in food a competitive edge and open the possibility for a more wide spread used of the new technique in Danish industry.

Collecting both the phase, dark field and the absorption terms in the X-waves introduces several new computational challenges. First of all algorithms for image reconstruction using phase are currently tested with only close-to-perfect signals, in an actual, and especially industrial, setting the current image-construction is not guaranteed to work. Even when a more robust methodology is used, the entropy in the phase signal is much smaller than in the known absorption signal and there will be a need to process these signals in real-time for any application outside a laboratory. Given that real-time signal processing for conventional CT technology is only now becoming a reality, the challenge of processing phase information in real time is a significant task.

The main focus of the strategic center is to provide state of the art solutions to the food production, but other research areas inevitably will benefit from the work by devoting a work package to dissemination of the developed technology. The center will invite Danish SME's and research communities to explore this emerging technology as an important part of the knowledge transfer. The applications are assumed to exploit the potential of the new imaging modality with respect to high sensitivity to light material composition, high spatial resolution and non-destructive measurements. The applicants will benefit from the synergic alliance between sensor physics, automated image analysis and high capacity computing founded in the proposed center.

The scientific conclusion and next steps

Over the last five years, phase contrast and dark field imaging have undergone a dramatic development witnessed by a range of high impact publications, and the prestigious Leibnitz price 2010 awarded to Prof. Franz Pfeiffer. The group at NBI has collaborated intensively with the Munich group by extending the range of applications in the medical and in the food area, including both phase contrast, and dark field imaging. Extending this collaboration to include a wider range of food products will give the Danish industry a unique opportunity to benefit from the technology before it is implemented by competitors. The collaboration will also train a number of PhD students who will be able to apply the technique to more industrial applications. The new X-ray imaging techniques have made significant impact in the scientific world and it is expected that new application and apparatus will emerge rapidly in particular in the medical area. However, applications for food products have as large a potential and Danish industry has the opportunity to be at the very forefront for the benefit of higher food quality and higher food safety.

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Animal genetic database

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Value for industry

- The establishment of a unique shared cattle breeding database.
- The implementation of a data collection and sharing system that eliminates duplication at farm and organization level.
- Development of a genetic evaluation system which identifies, on a worldwide basis, those cattle that are most profitable under Irish conditions.
- The animal genetic database help supporting and promoting increased international collaboration in beef breeding, animal health, and genomics.

Background

Over the last thirteen years Ireland has established a new infrastructure to facilitate the genetic improvement of both dairy and beef cattle. Prime responsibility for leading the development rests with the Irish Cattle Breeding Federation Society Ltd (ICBF) established in 1998 with the objective of achieving the greatest possible genetic improvement in the national cattle herd for the benefit of Irish farmers, the dairy and beef industries and members. This development has been funded by a unique partnership involving farmers, breeders, service providers, service income and Government.

Why work is needed

The focus of this paper is on the developments that have impacted on the availability of data for use in creating information essential for effective cattle breeding decisions. These have especially included: creation of the ICBF Cattle Breeding Database, implementation of the Animal Events (AE) data collection system, and the creation of linkages with other data collection systems.

The methods used

At the time ICBF was formed there were a large number of separate computer systems supporting aspects of cattle breeding in Ireland. Each had its own data collection system and supported the information needs of one or other aspect of the cattle breeding industry.

For example, each Herd Book (there were 18 at that time) had their own system, each Milk Recording organization (there were 8 in 1998) had its own system, and Department of Agriculture, Food and Marine (DAFM) operated separate systems for genetic evaluations and the official calf registration and cattle movement monitoring system (AIM-CMMS). These systems used several different animal identifications and held limited cross-references.

Creating the database involved an enormous effort to: negotiate agreements for the sharing of data, to establish shared data collection systems and to consolidate the existing computer files into a single shared database. The key principles underpinning the agreement between organizations to share data are summarized in table 1.

Table 1. Principles of data and information sharing agreement underpinning ICBF database.

No.	Principle
1.	Contributors of data to the creation of the database retain “ownership” and can obtain a copy of their data at any time.
2.	All data originating on farm, and known first to the farmer, is captured through “Animal Events” a system controlled by ICBF.
3.	ICBF operates an industry wide network of systems to facilitate the electronic sharing of relevant data collected for other purposes. Examples include; inseminations, slaughter data, and sale data.
4.	All data in the database is available for research subject to a minimal set of conditions.
5.	Genetic evaluations are an integral element of the database.
6.	Herd owner’s control service provider access to herd and animal data.
7.	Service providers have access to data and information systems needed by their particular businesses for those herds that have granted access.
8.	HerdPlus® is a service provided by ICBF to the herd-owner that facilitates access to all data and information relevant to the herd in the database.
9.	Service fees are set on the basis of User Pays and Full Cost Recovery.

The ICBF cattle breeding database supports, through the use of a range of new technologies (Cromie *et al.*, 2008), the information needs of milk recording, herd books, AI organizations and cattle farmers. Farmers are able to access their own data in the database through the HerdPlus® web service. Figure 1 illustrates the data sources, information outputs and services that are currently supported by this database.

It is important to note that Genetic Evaluations are a peripheral yet integral element of the ICBF database. All data used in the evaluations is sourced from the database and all results returned to the database from whence they are published and distributed.

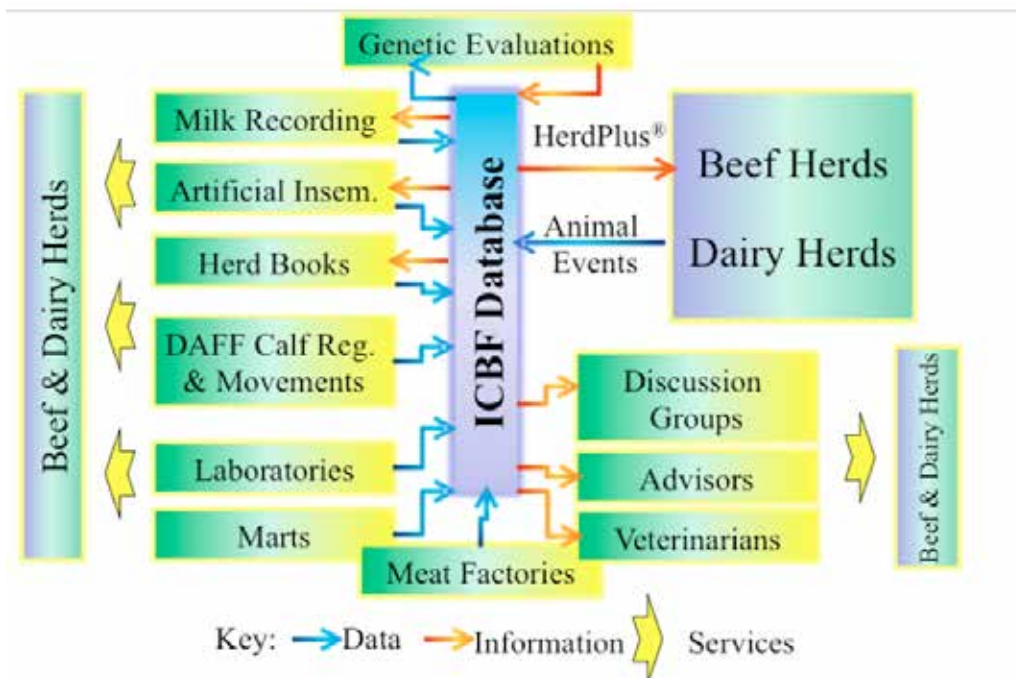
The AE data collection system was developed, as part of the overall database development, to replace the overlapping data collection systems operating in 1998. This system was built to remove duplication in data collection, at farm and organization levels, and to ensure all the data required for cattle breeding and other official purposes was collected efficiently and accurately.

The AE system collects data on those cattle breeding events, e.g. calving, birth, identification, mating, ..., which are first known to the farmer.

Both paper and electronic systems are supported. The data collected in this way is accessible to those participating organizations that provide cattle breeding services to the herd.

The ICBF database has been fully operational for dairy, beef, milk recording, beef performance recording, genetic evaluations and herd books since 2005. Some 81,000 herds, with 1.8 million calvings, representing ninety percent of the entire Irish cattle herd, were participating in one or more aspects of the database by the end of 2011.

Figure 1. ICBF database showing data sources, information outputs and services to farmers.



The ICBF database has access to data collected by a wide range of organizations for other purposes. The data collected and stored in the ICBF database from these other sources, includes:

- Calf registrations through DAFM – all calves born in Ireland are first registered by DAFM, and only then added to the ICBF database. This ensures the official EU identification is available for all calves entering the ICBF database.
- Cattle movements, exports and deaths through AIM-CMMS ; this eliminates the need for any of the cattle breeding organizations to collect this data. A nightly data feed is provided to ICBF for all movements into or out of herds participating in the database.
- Slaughter data from meat processing plants in Ireland : slaughter date, carcass weight, carcass grade, fat score and, the two images used in carcass grading.
- Sale data from auction marts. This includes dates, weights albeit not always for single animals, and prices.
- Milk records from Milk Recording organizations. The ICBF database is an integral part of the milk recording and result reporting process that operates in Ireland.
- Artificial inseminations recorded by technicians. ICBF has developed a hand-held computer system that links directly to the ICBF database for insemination recording. This system is used by all the main AI field service companies operating in Ireland.
- Linear scoring, dairy and beef, and weight recording services. The same handheld technology used for AI technicians is provided by ICBF for linear scoring and classification services.
- Farmer recording through dedicated web screens (weights etc).
- Pedigree registrations: production of pedigree certificates and DNA labels.
- Genomic services: recording of tissue samples for DNA extractions.
- Health monitoring linked with Animal Health Ireland.

These linkages ensure no redundancy of data recording. The result is a greatly increased availability of data to all participants in the ICBF database.

The scientific conclusions

In the last thirteen years the Irish cattle breeding industry has undergone a complete redevelopment of its data gathering and genetic evaluation infrastructure. The key developments include:

- The establishment of ICBF as a working partnership between the organizations involved in Irish cattle breeding
- The establishment of a shared cattle breeding database
- The implementation of a data collection and sharing system that eliminates duplication at farm and organization level
- Development of a genetic evaluation system which identifies, on a worldwide basis, those cattle that are most profitable under Irish conditions
- Supporting and promoting increased international collaboration in beef breeding, animal health, and genomics.

Irish farmers, research scientists, Herd Books and AI Companies have responded by making good use of the greatly increased amount of information now available. As a result Irish farmers are now able to better exploit the potential of genetics as a tool for improving the profitability of their enterprises.

The next steps

The ICBF database will continue developing and adapting to new context for the benefit of the whole Irish farming industry (e.g., mobile technology).

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Use of biomarkers as tools for tracking and tracing meat and meat products and to predict and monitor meat quality

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Value for industry

- Integration of the production chain. Increased speed of the production process through the selection of carcasses for meat quality shortly after slaughtering.
- Monitor traits such as recovery from transport stress and the related meat quality of slaughter pigs in the slaughterhouse.
- Prediction of the final meat quality shortly after slaughtering.
- Measurement of the potential of an animal for a trait (genotype to phenotype transition).
- Tracking and tracing such as proof of the management conditions during the life of the animals. For example, proof of the outdoor husbandry of dairy cows based upon the product composition in relation to the feed differences: fresh grass outdoor vs hay indoors.

Background

Biomarkers are tools to predict, monitor, and manage physiologic processes and (economic) traits. They can be used in human medicine, but also in science and industry. An example of industrial use is to predict meat quality either while the animal is still alive or directly after slaughter. With this knowledge the meat quality can be modulated by breeding or in the slaughterhouse by interaction with the ageing process. Scientific use is for elucidation of the biological processes underlying the development of meat quality, both during life and post mortem.

Today genomic DNA markers are increasingly used in animal breeding. Current technologies enable the use of many – up to several hundred-thousand – of such genetic DNA markers. Biomarkers go one step further since they are the result of the interaction between the genome and the environment. Thus, biomarkers can differ in time and location. Biomarkers can potentially detect whether an animal

experienced changing conditions such as housing, feed, relating to management and global position. Thus, biomarkers have important properties for detecting authenticity and tracking and tracing of food products, and for predicting related product quality parameters.

Several (omics) technologies enable the development and detection of biomarkers. We have experience in using transcriptomics and proteomics, and starting up metabolomics technologies for the development of biomarkers. Using these technologies we first unravel the underlying biological mechanism using bioinformatics and advanced statistics. The use of systems biology may further show the body-wide effects of the biological mechanism. With this information we develop fast and easy to run biomarkers test methods, and validate these in an independent group of animals.

Why work is needed

During the past decade genome sequencing technique developments enabled the fast and cheap sequencing of individual genomes. Analyses of these found an increasing number of genetic differences between individuals, called Single Nucleotide Polymorphisms (SNP). Nowadays for an increasing number of livestock species SNP-CHIPS with tens of thousands to hundreds of thousands SNPs are available.

This speeds up the search for genetic differences between traits. However, many production traits - including especially meat quality traits - are only partially genetic determined. Some traits even have a low genetic component, and most of the phenotype is influenced by environmental factors, for example the feed of the animal, the management (e.g. housing, stress, etc.) of the animal, temperature of the environment, etc. Such traits are difficult to measure using genetic markers. Biomarkers do not measure static differences between genomes, but dynamic differences between the expressions of the genomes of individuals.

The dynamics can be influenced by all the environmental parameters. Thus, biomarkers provide a better representation of the present status and developments of the trait than genetic markers. Therefore, biomarkers can be used to monitor a trait. If the biological process underlying a trait is known biomarkers can also be used to predict the final outcome of a trait. During our work we found that we could predict several meat quality parameters with good accuracy using our biomarkers. Similar results are achieved in biomedicine where biomarkers are increasingly used to indicate biological functioning of organs in several metabolic diseases.

The methods used

The development of a biomarker is a three step procedure. In the first step the need for a biomarker is expressed, e.g. by an industry. In a first screen the literature is searched for existing knowledge of the underlying biological mechanism. When this knowledge is lacking in the second step biological samples expressing the trait of interest are investigated using functional genomics technologies (transcriptomics, proteomics, and metabolomics).

Differences in the expression profiles correlating with differences in the phenotype of the trait may be the first indications of a useful biomarker. During this phase bioinformatics, statistical evaluation, and systems biology methods are used.

Validation of the results using independent samples and - if possible - independent technical methodologies are indispensable. If a biomarker is confirmed and validated the final step is to prove functionality in an implementation phase. Especially in the latter phase innovative methods are required to measure the biomarker on site, in line, real time. Technological innovative SMEs are presently developing such (fully automated) tools. Furthermore, measuring the biomarkers without laboratory skills comes within reach.

The results obtained

Biomarkers are molecules indicating the present status of a biological process. Biomarkers need to be quantitatively measurable. The biomarkers were developed using transcriptomics and proteomics techniques, analysed using bioinformatics and statistical methods, and validated using independent other pork production chains and techniques. The results show that especially meat colour, ultimate pH and drip loss can be predicted based upon biomarkers. For the latter two meat quality parameters we obtained an average predictability of about 50%, but with a maximum predictability of up to 80% using a combination of 20 individual biomarkers, each with a small effect, but with a higher predictability if combined. This means that when measuring biomarkers and later verification with real measured drip loss and ultimate pH the biomarkers were correct in 80% of the samples.

Transport of slaughter pigs to the slaughterhouse is stressful for the pigs and stress deteriorates meat quality. While it is known that resting the pigs restores meat quality the optimum length of the resting period is unknown. In an experiment pigs were exercised on a treadmill for approx. 30 minutes as a model for transport stress, and rested afterwards for 0, 1, or 3 hours. The results were compared with unstressed pigs. Proteomics of muscle samples showed that several biomarkers indicated simultaneously the effect of stress (exercise), resting time, and meat quality. Monitoring the expression level of biomarkers such as those indicating stress, resting time, and drip loss for example can be help to optimise resting time and meat quality.

The scientific conclusions

Biomarkers can highlight the biological mechanisms underlying all production traits. Knowing the biological mechanisms provides the possibility to monitor a trait by measuring the status of the biological mechanism via measuring the expression of the molecules involved. Furthermore, it offers the possibility to change the trait by changing the expression of the biological mechanism. There are multiple ways to do so, e.g. breeding changes the genetic capacity for a trait, nutrition changes the expression of genes that changes the expression of the biological mechanism in which these genes act, etc. Especially the latter way of affecting a trait requires the knowledge of the underlying biological mechanism.

The next steps

A number of research project during the past decade focussed on developing biomarkers for an increasing number of traits. As a result of this the number of biomarkers available has increased. However, quite often good validation of biomarkers is lacking due to the necessity of using (large) independent sets of samples with adequate phenotypic measurements of the traits, which is expensive. For a limited number of biomarkers these requirements have been fulfilled, and the first experiments aiming to implement the biomarkers are on the way. For the near future validating and implementing of biomarkers will be important to prove their importance for the livestock industry.

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Individual animal traceability from farm to boning room - A case study

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Background and introduction

Silver Fern Farms (SFF) is a Farmer co-operative with 20,000 suppliers, 22 sites (17 slaughter) - Beef, Venison, Lamb with a turnover of €1.2b. We process 30% of all Beef, Sheep, Lamb & Deer in NZ.

Opportunities for improvement in the value chain lie on the farm through improved productivity, with better feedback of processing information from plant to supplier leading to improvements on farm and at the processor. The key to this is moving the focus to individual animals & understanding the variability. The main focus for the last few years has been the PGP “Primary Growth Partnership” project. PGP is a government-led strategy for the whole primary production sector (Figure 1).

SFF joined up with Landcorp (largest farming company in NZ- government owned) to successfully propose a PGP project worth total of €90M over 7 years (45% of that from SFF). FarmIQ Ltd. is the company formed to run the project and to develop the database to collect all the data. The project calls on outside expertise for R&D.

Traceability as an enabler of other technologies:

SmartStim - Intelligent electrical stimulation (developed by Carne Technologies)

- Electrical stimulation of carcasses post dressing applied whilst the carcass is on a load cell. The load cell measures the muscle response to the electrical input, and predicts the ultimate pH of the carcass.
- Electrodes in the stimulation tunnel are individually controlled to control the ultimate pH.
- Carcass tracking essential for this and RFID readers are used at the entrance of the stimulation tunnel.

Figure 1. The structure of the primary growth partnership programme, that has an investment of €90M over 7 years (45% of that from SFF).



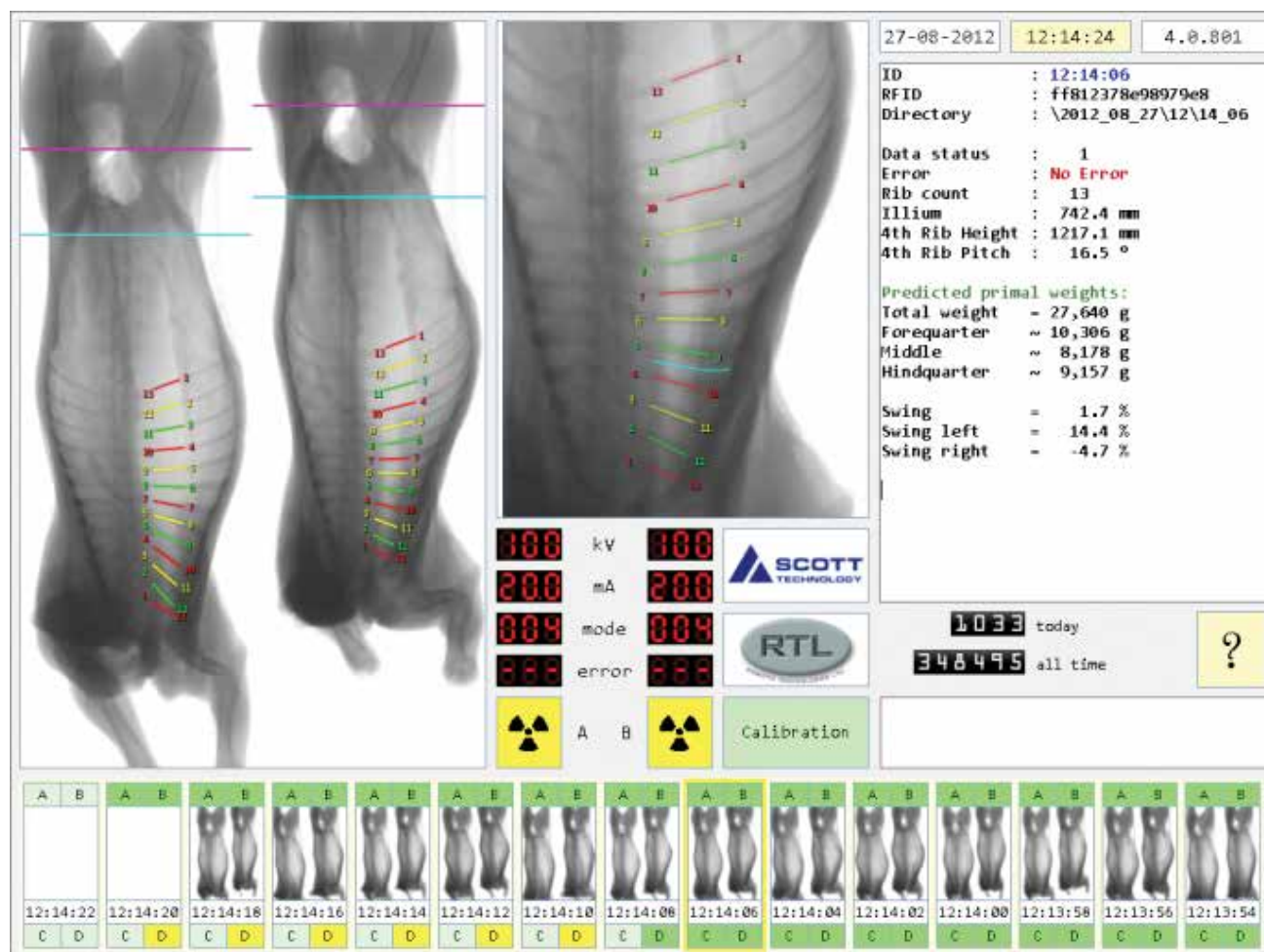
The lamb primal cutting machine needs objective measurement because the cut positions vary according to the carcass conformation. Rib position and angle needs to be ascertained by X-Ray (Figure 2). All these data collected can be fed back into the carcass database because of RFID skid tracking. We are looking at DEXA development for meat/fat/bone proportions. We are X-Raying and analysing in real time at chain speeds of 30/minute (two X-rays). RTL robotic technologies Ltd is joint venture between SFF and Scott Technologies.

Collaboration is underway with AgResearch on near infrared (NIR) spectroscopy for beef and with Massey University on lamb. Agresearch are applying NIR on pre rigour beef within 60 minutes

of slaughter to predict ultimate pH, colour and tenderness. The Massey work is on lamb loins 24 hours after slaughter for input to genetics programs. RFID trays are being trialled to measure yield of individual cuts from individual animals. Cuts can be related back to animal ID. This is a trial set up in one of our development rooms, and is a joint venture with SCL Ltd. A Marel cut tracing system has also been commissioned at the new TeAroha beef plant to compare with the tray-based system.

Silver Fern Farms are not convinced that there is a business case to trace cuts to market, because carcass identity is lost downstream and because there is currently no regulatory requirement or sufficient customer demand.

Figure 2. A screenshot of the primal cutting machine that uses X-ray to determine the position and blade angle of robotic primal cutters that split the carcass into legs, middles and forequarters.



Where does all the data end up?

Farm IQ is the company set up to create a demand-driven integrated value chain for red meat that delivers sustainable benefits to all participants: farmers, processors and marketers. It brings all the data together into one database. Farmers can cut & dice the data in many ways to look at all aspects. The on-line interface is working now with our 400 pilot farms, and will expand up to 7000 farms and beyond.

NAIT - National Animal Identification & Tracing. NAIT Ltd is an industry-owned government sponsored non-profit company implementing the NAIT scheme. It is mandatory for cattle (1st July 2012) & Deer (1st March 2013).

NAIT maintain database recording all animals, location, PICAs & movements. Every animal has an ID - tagged at birth. Every property has an ID (Linked to Ministry of Primary Industry "farms-on-line" property mapping system").

It is primarily for movement tracking for quick response in the case of a biosecurity threat such as disease outbreak & for continued market access. It is a system for recording physical movements and is built around the EID tag.

Traceability in the processing plant: Beef

On farm, all farm metrics (weight gains, forage, genetics etc.) sent to Farm IQ are indexed by RFID tag. Origin and destination information is required by NAIT for all movements to the slaughter plant via sale yards.

At truck unload, the tags are read and animals (and tags) are sorted into herds. After slaughter as carcasses join the slaughter line, the tag is read again and linked to RFID trolley or skid. The chain is interlocked so that every tag must be read or manually entered. Along chain there are RFID trolley readers at legger & grader points. As the carcass moves out of the chiller and into the boning room, it moves over a load cell with an imbedded reader. The cold carcass weight is recorded for boning room yield calculations & weight loss statistics.

Traceability in the processing plant: Deer

Deer carcasses yield information is determined using a weigh rail boning technique. Deer primals are usually taken off the carcass in the same order every time. This allows cuts to be weighed by subtraction while the carcass is hanging on the rail as each scale has an imbedded RFID reader. The system is still under development, but there are plans to implement primal weight recording at all deer plants in the near future. Initial trials were based on manual tracking, but were unsuccessful, RFID solved this problem.

Traceability in the processing plant: Sheep, lamb & calves

Currently (2012) EID is not mandatory for sheep in New Zealand, but EID tags are issued by Silver Fern Farms to participating farmers who wish to make use of EID technology. Farmers enter on-farm data (e.g. genetics, age, breed, irrigation, fertiliser, weight gains, forage types etc.) that are linked to the tag ID.

Upon arrival at the processing facility

The first sheep in mob has a high-frequency RFID tag attached, after scanning in yard. After slaughter, sheep EID ear tags are scanned read on the chain position (two readers to cope with breakdowns or misses). After scanning the tag is removed as the carcass progresses along the process line. The mob number & data are assigned to the slaughter line position at this point. Data are in a shift register to simulate the real chain. It was found that when RFID readers are on a spreader, the height range is small (300mm) which enabled a smaller antenna that produced less noise to be used.

As the carcasses progress along the processing line, all attachments move in unison (Synchronized) - indexing the proximity moves all data, right along the chain, two readers are used at the inspection station and at the weigh scale to avoid misreads. Data in a shift register to simulate the real chain. For detained carcasses, once they pass the detain switch, data is in a virtual "bucket". As carcasses enter the boning room, the RFID skid is read, the weight recorded, and the validity of that item number is checked. Carcasses are grouped into batches, and electronic documentation is recorded from carcasses that have come from off-site.

Equipment selection: EID tag readers

Tag readers from two manufacturers, one Australian, one NZ – Aleis & SCL have been trialled and displayed similar performance.

EID Tags: EID tags for Beef & Deer are defined by NAIT. There are three main manufacturers in NZ, all tags must be approved by NAIT. There are several numbering schemes and several formats which makes it difficult for manual entry. The same tags were selected for sheep so the existing readers could be used.

Skids: Existing skids which was on the market were incompatible with existing systems at processing plants, and due to their construction would rapidly break and wear. The challenge was to insert tags into existing skids which would work while surrounded by steel, in an electrically noisy environment. A solution was found in collaboration with SCL Ltd. Where skids were punched to create a hole for the tag, then tags were injection moulded into the skids. Injection moulding is done on site with our own staff so that production is not affected. The total price to tag a skid is around €5 each. Some plants have up to 45000 skids; examples are shown in Figure 3.

Control equipment

20 years ago the company decided to use industrial electronic equipment based on programmable logic controllers (PLCs) and industrial touch screens rather than PC-based solutions for carcass grading systems and data input. This philosophy has been expanded to incorporate traceability inputs. The system uses Rockwell PLCs with distributed I/O and Rockwell touch screens running on Ethernet/IP which is very reliable; few maintenance problems are encountered as because technicians are very familiar with the technology. The system is very fast and there are plenty of interface options for scanners, scales, printers, touch screens, RF readers, pin stops, chain interlocks etc. All software is developed in-house, so any required changes can be made very rapidly.

Skid databases

Because there are large numbers of sheep and lambs being processed, a PC-based SQL database is used. The PLC will query database over a network, and data is typically returned in 0.2 seconds. Speed is important for real time actions such as rail switching for carcass sorting etc.

There are smaller numbers of cattle and deer being slaughtered so data can be held in PLC (hot bone beef plant only has 350 skids). This system has a millisecond response time and no reliance on a commercial network. A further advantage is that the skid ID does not need to be known by any other system.

Challenges:

- Never attain a 100% read rate, 99.95% on EID skids.
- Ear tags – sometimes damaged but usually because of incorrect application.
- If two skids are on one attachment, cannot read either skid.
- Most of our installations are surrounded by steel.
- Tag die-off, a high rate at first then slows but still 0.5%.
- Interference – electrical noise – especially VSDs and electronic ballast fluorescents plus other readers in the area (portable wands or other fixed readers) and even other tags (in pockets etc.).
- The larger the antenna, the more susceptible it is to noise. Antennas must be large for beef and deer, SCL have developed an antenna comprising multiple small antennas, multiplexed.
- Skid speed - manual verses mechanised chain.

Figure 3. RFID skids that can be used to main carcass traceability.



Short overview of electronic identification in bovines, and prospects for alternative transponder technologies

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Value for industry

- There are a range of Electronic Identification (EID) options for the industry, and it is important to evaluate the strengths and weaknesses of each option.
- Scotland's Rural College are currently evaluating an alternative ultra-high frequency option with other commercial and academic partners to establish the potential performance benefits.

Background and introduction

Electronic Identification (EID) ear tag application has been a requirement in the sheep sector since January 2010 under EC 21-2004. The standards adopted by EC 21-2004 are ISO 11784 and ISO 11785. These specify both the technical concept for radio frequency (RF) identification (ISO 11785) and also the structure of the radio-frequency identification code for sheep (ISO11784). These are Low Frequency (LF) inductively coupled devices operating at 134.2 KHz. All passive EID devices use RF energy to excite the transponder. This can limit the range at which they may be read, especially inductively coupled devices.

A number of application areas do utilise LF RFID effectively in cattle farming, such as automatic weighing, shedding, milking and automated recording of individual feed intake and other close range systems. Apart from the benefits which can be gained from EID for cattle on-farm management there is also a considerable interest across the world on the question compulsory EID tagging for the cattle sector. For example, New Zealand introduced the mandatory electronic Identification using LF RFID in cattle on the 1st of July 2012 (Nason, 2011). Other countries already operating a mandatory system are e.g. Australia (Nason, 2011), Canada (Sundermann and Pugh, 2008), Uruguay (Swedberg, 2008) and Denmark (Swedberg, 2012).

However, newer alternative ultra-high frequency (UHF) transponder technologies are showing significant promise. They are used in other industries, and commercial implementations for bovines are starting to appear on the market.

UHF Technology

UHF technology has improved significantly within the last 10 years, becoming a useful technology, and adopted broadly for tracking goods in the supply chain (Pugh, 2004). It also became an interesting technology to be considered for livestock purposes. New Zealand, although opting for the LF option for their cattle, conducted trials to investigate the early application of UHF RFID technology for animal tagging looking at three different species, including cattle (Sundermann and Pugh, 2008).

UHF ear tags potentially have a number of advantages. The read-range is much higher (in the order of metres read compared to decimetres in LF RFID). This makes UHF more adaptable to reading off the identifications at commercial locations such as auction marts, abattoirs, animal transport vehicles etc. The UHF transponders themselves are capable of storing information as opposed to just containing a unique identification code, and data transfer rates are orders of magnitude faster.

UHF technology implements anti-collision as standard which means that multiple tags can be read simultaneously, for instance a batch of animals passing under through an opening would not require singulation to be read effectively.

The antenna reader devices can also write information to these devices when an animal passes within range. So in future, management information could be applied and stored on individual animal tags, and subsequently retrieved directly from the transponder. Data such as recorded weight,

medications applied, movement history, breed, sex and date of birth etc., could all be potentially stored on the tag, and conveniently retrieved where appropriate. SRUC is currently conducting a trial to validate performance of the read/write capability in farm and other situations.

There are also appropriate UHF handheld readers available on the market which can be very useful for on-farm management purposes to read the UHF ear tags and also to write to them. However, these handheld readers are not specifically made for the agricultural market but for other industrial supply chains.

EID technology is moving fast, and new opportunities will arise due to increasing performance and reduced cost for UHF transponders and reading devices. The advantage of this technology is that it is used so broadly across different industries that a fast developments and implementations may be anticipated.

In Scotland, ScotEID (www.scoteid.com) runs an extensive field study of electronic tagging and under the current Phase III of the pilot project, an evaluative field testing study of UHF tags alongside LF equipment in cattle is currently underway (Moxey, 2011). Moxey states that inclusion of UHF technology in the pilot project is because UHF systems are capable of accurately reading multiple tags at a much faster rate and greater range.

There are already two USDA approved UHF cattle tags commercially available on the market (Eriginate's eTattoo tag and Hana Innosys tag; Figures 1 a and b). Figure 1 b shows the eTattoo tags, which have the same size and shape as a conventional Size 5 cattle tag. The Hana tag has a slightly different shape.

Figures 1 a and b. Dairy cows tagged in the right ear with Hana Innosys UHF tag (a) and Eriginate's eTattoo ear tag (b).



The eTattoo ear tag was tested over a period of 6 months (Dairy White Paper, 2010) and the findings showed that the tags with a Sirit reader system can achieve greater than 99% tag capture in a 19 foot alleyway with 4 antennas placed 12 feet overhead. However, the report states that body masses may have more easily blocked a successful read of a tag and these findings are important and indicate that cattle behaviour can also affect the ability to successfully capture the tag ID and not simply size of the reading zone.

Brazil is also trialling UHF technology for the use on cattle and water buffalos (Swedberg, 2010) using Eriginate's eTattoo dangle tag. The water buffalos were specifically chosen to test the tag's durability and they performed well, as reported by Swedberg (2010).

The scientific conclusions and next steps

Overall, when reviewing literature on bovine EID, useful applications of electronic tagging can be found and are more and more used on farms. A number of countries have already made electronic tagging compulsory. Those countries usually opted for the LF technology. The already high number of LF ear tags and boluses in the livestock sector might be seen as a stumbling block for the introduction of UHF. However, both technologies can easily be operated along side each other because there are no electromagnetic issues running them in parallel.

The UHF technology is developing fast on a global scale across a various industries which give the technology an innovative momentum. The advantages of UHF ear tags make them certainly interesting for the bovine sector and the few reports which are already published shine a very favourable light on the technology so far.

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Pigtracker – UHF-Based RFID Ear-Tags for Pigs

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Value for industry

- UHF-RFID is useful in pig breeding and production.
- The reading distance is approximately 2 meters.
- GS1 is the best data standard for UHF-RFID.
- There are still some challenges for reading RFID at the slaughterhouse.

Background and introduction

Currently, most cattle in Denmark are tagged with low-frequency (LF) ear tags, as is required by federal mandates. Meanwhile, farmers are not required to track their pigs electronically, but many have also tried the LF tags in order to improve their visibility into each pig's whereabouts, and to provide an automated record of every animal's history. With LF tags, it is not possible to read many pigs at the same time or read tags from a distance. UHF technology makes it possible to read tags simultaneously at a greater distance.

The primary goal of the project was to develop the ear tag for use in pigs and to test both stationary readers and handheld scanners under normal production settings at the farm and in the slaughterhouse.

Additional goals were also to find the best RFID standard for identification, and develop the IT-system for data handling and tracking.

Why work is needed

Through setting up a system for identification and tracking of pigs, the eventual profit will depend on to what extent the data generated by the system, will be utilized in the different parts of the production process from birth to slaughter. Individual pig data could be: birth place, removals (date and physical location), medical treatments, production traits, slaughter quality etc.

The methods used

Five breeders were involved in the farm testing and one slaughterhouse. One of the most important points in the project was to develop a new ear tag based on UHF RFID, which will be necessary for:

- Multiple reading.
- Longer reading distance > 5 meters.

Reading at the farm was tested both with antenna at the wall, at the ceiling and a handheld antenna for individual reading. The ear tag used in the test is shown in Figure 1.

The partners in PigTracker are: Pig Research Centre – Danish Agriculture & Food Council, Prosign RFID, RF-Labeltech and DMRI- Danish Technological Institute.

PigTracker has received support from the Danish Ministry of Food, Agriculture and Fisheries.

For more reading see the projects homepage: www.pigtracker.dk

Figure 1. The developed eartag.

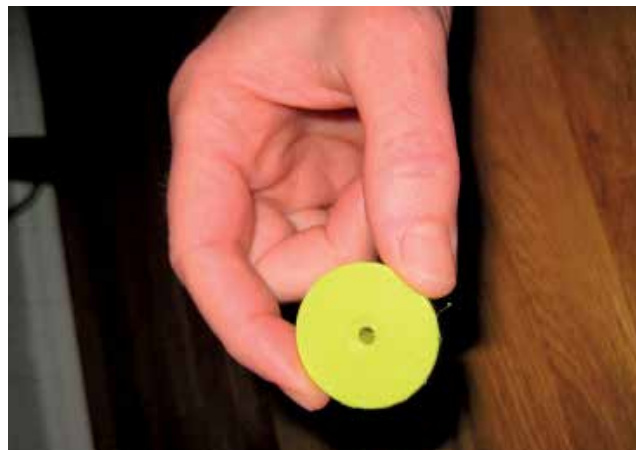


Figure 2. Reading the ear tags after scraping at the abattoir.



Figure 3. Dirty eartags can be difficult to read.



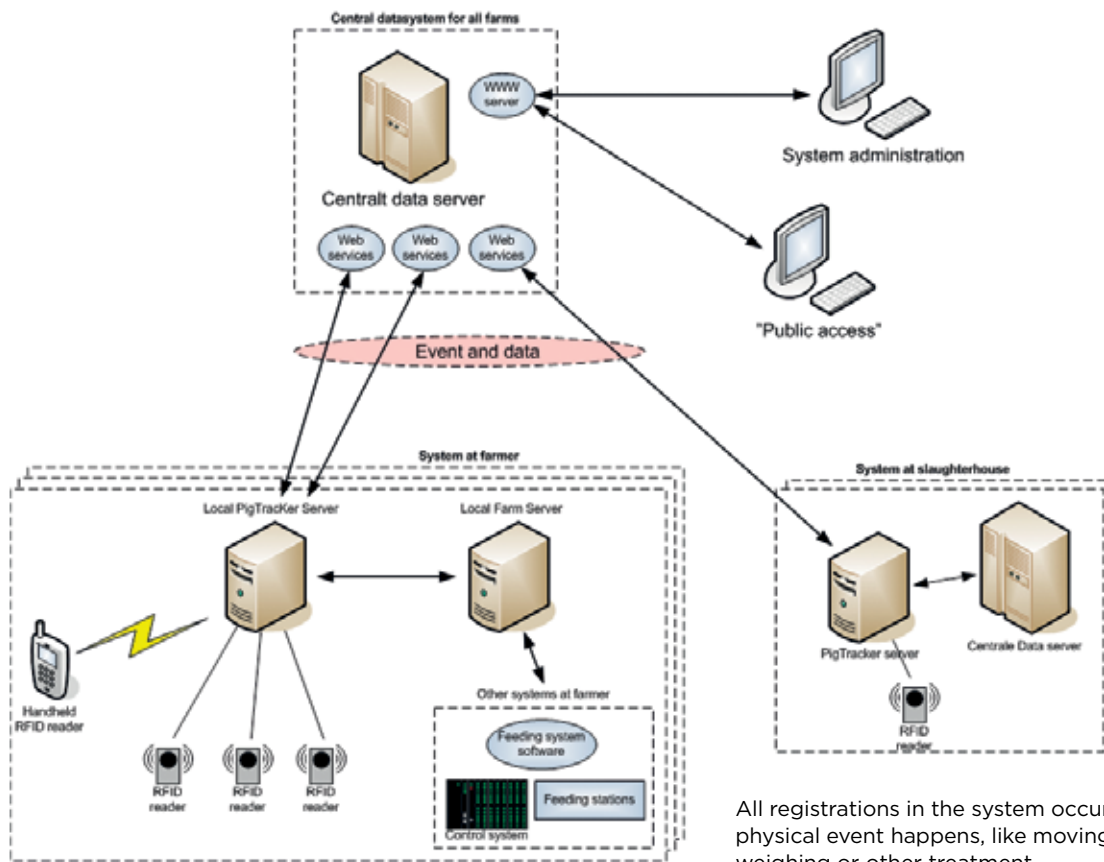
The results

The reading distances during field test at the farms were up to 2.5 meters with the stationary reader and not more than 1 meter with a handheld reader. When the ear-tag is clean it is easy to read and the reading skills are close to 100%. The reading ability was reduced with dirty tags, and moreover it is necessary with air between the tags and the reader, because ultra-high frequency radio frequency is not capable to penetrate body tissue.

The investigation at the slaughterhouse showed it is more complicated to use the UHF tags in this environment. At arrival it is very difficult to read due to all the steel in the area, and it was also very complicated to find a place to mount the antenna in a Danish abattoir.

The readers were placed just after de-hairing (Figure 2) to combine the ear-tag to the slaughter ID at the gambrel. 5-10% of the tags was loose during the slaughter process and due to the UHF tags it is more complicated to be sure to read the right tags. Some of the tags are not able to read due to water inside and the antenna must be in plastic box to protect under cleaning. This will also lower the reading distance.

Figure 4. Dataflow and sequence



All registrations in the system occur when a physical event happens, like moving the pigs, weighing or other treatment.

Figure 4: The IT system structure and the different stakeholder's part in the system:

- *Storage of data from each farm*
- *Regulatory requirements in relation to the tracing, hazard, environment*
- *Trade in live animals between producers in Denmark and for export*
- *Communication between producer and slaughterhouse*

IT System structure

PigTracker used the GS1 EPC standards for the unique identification of the pig and the location, and for the exchange of data between the reader and the central server, instead of the existing ISO 11784- and ISO 11785-standards, which currently are the standards for livestock identification.

This system:

- Considers each pig as a individual trade item → SGTIN.
- Considers each farmer as a location → GLN.
- Considers each herd and lairage as a serialized number within the location → SGLN.
- Will be able to handle approximately 268 million individual numbers.

The scientific conclusions and next steps

UHF-RFID is useful in pig breeding and production and special for the pig producer it is easy to know the numbers and placement at the pigs in each farm. The reading distance is approximately 2 meters and the best place of the antenna will be in the ceiling over the area where the pigs had to be moved from one place to another. GS1 is the best data standard for UHF-RFID, and is already a well-known standard in other industries. There are still some challenges for reading at the slaughterhouse, and to use the UHF technology in the best way.



Notes

Lined area for notes, consisting of multiple horizontal lines.

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