Rapid prediction of pork quality

Correlation of fresh meat measurements to pork water holding capacity and its technological quality

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Abstract

Water holding capacity (WHC) of pork defines the sensory appreciation and processing yields of meat. Pork varies in WHC and is mainly generated by differences in post mortem muscle metabolism of carcasses. Nowadays, the pork processing industry performs sorting of carcasses and primal cuts on the basis of weight and lean characteristics. Additional sorting by WHC can further optimize processing yields of pork products.

The aim of this thesis was to validate rapid prediction of pork WHC. The first objective of this thesis was to investigate the possibilities of a rapid prediction of pork WHC by measuring parameters such as pH, colour L*, drip loss%, water absorption, and by NIRS at laboratory scale and at pig processing plant scale. Results revealed that NIRS prediction equations could be developed to predict drip loss% and colour L* of pork samples. Equations for colour a*, b*, and pHu were not applicable for prediction of WHC.

The positive results of NIRS to predict WHC and colour L* at laboratory scale led to further research to study NIRS prediction of pork quality (pH, colour L*, and WHC) under pig processing plant conditions. It was concluded that NIRS prediction equations can be used for screening WHC at pig processing plants.

Also, characterization of moisture loss from muscle early post mortem and whether these losses are useful in predicting WHC of fresh pork was investigated. Results revealed moisture losses from muscle tissue early post mortem which suggested that select time periods correspond to culmination of biochemical and physical events facilitating moisture release, which can be used for early drip prediction. Results suggested an approach for capturing moisture release early post mortem which may be used to predict WHC in pork.

The second objective was to investigate if predictions of pork WHC could be used to optimize processing of pork. Technological yields could not be predicted ($R^2 < 0.21$ and RPD < 1.1) by NIRS. Pre-selection of back bacons by NIRS predicted WHC values, did result in significant different average pHu and colour L* between both groups. It was concluded that NIRS can be used to predict rapid fresh ham quality for sorting and optimization of the cooked ham process.

The overall conclusion of this thesis is that NIRS prediction equations for WHC can be developed for pork loin samples measured at pig processing plants and that these prediction equations can be used to optimize processing of pork.

Voorwoord

De specifieke behoefte om de vleesverwerking verder te optimaliseren heeft geleid tot het ontstaan van dit proefschrift. VION Food Group en Nutricontrol hebben samen deze behoefte gesignaleerd en een promotietraject gecreëerd. Toen ik hoorde van het onderzoeksproject om op een snelle manier vleeskwaliteit te voorspellen was ik meteen enthousiast.

Voor mij is het erg belangrijk dat de waardevolle producten van deze sector maximaal benut kunnen worden. Tijdens mijn studie heb ik mij voornamelijk met runderen bezig gehouden en had mij nooit erg in varkens verdiept. Aan het begin van mijn promotietraject wist ik daarom heel weinig van varkens en varkensvlees. Gaandeweg heb ik geleerd dat de varkenssector veel dynamischer en complexer is dan ik mij ooit had kunnen voorstellen.

Ik heb met heel veel plezier als promovendus gewerkt, met dit proefschrift als tastbaar resultaat. Het plezier en het eindresultaat zijn mede tot stand gekomen dankzij de betrokkenheid en het enthousiasme van een groot aantal personen. Graag wil ik een aantal personen daarom ook in het bijzonder bedanken voor hun bijdrage.

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Chris

Opgedragen aan mijn oma Klaasen

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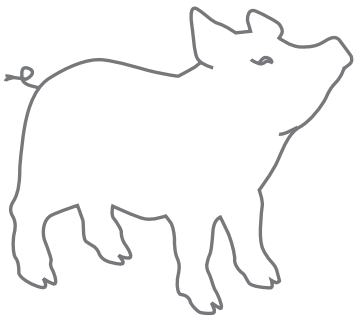

General Introduction

1. Introduction

Pork is the major meat consumed in the world (FAO, 2012), and is estimated to 99 million tons per year (Table 1). The Netherlands is responsible for 0.5% of the world pork production (FAO, 2012). Within The Netherlands, 16.7 million inhabitants consume on average 40.7 kg of pork per person per year (CBS, 2012).

1.1 Morphology of pig muscles

After slaughter, pig muscles convert into pork. The peri mortem physiology of pork is of utmost importance during the development of pork quality. Muscles are attached to a bone by a tendon which is attached to the epimysium. The epimysium covers the entire muscle consisting of several muscle bundles and fibers (Figure 1). A bundle of muscle fibers is covered by endomysium and perimysium which connects muscle fiber bundles. The epimysium, endomysium, and perimysium are connective tissues in which blood vessels and lymph vessels can be found.

Table 1. Pork production in million tons per year and percentage per country/region (FAOSTAT, 2007).

Country/region	Quantity (million tons) 2007	%
China	44.02	44.4
United States of America	9.16	9.2
Europe	25.97	26.2
The Netherlands	0.54	0.5
Other parts of the world	19.36	19.5
World total	99.04	100

A muscle fiber is a formation of myofibrils that consist of thick (myosin) and thin (actin) filaments, which are arranged in sarcomeres (see Figure 1). A myofibril consists of multiple sarcomeres longitudinal alingend next to each other. The thin filaments within a sarcomere are connected to Z-discs or Z-lines, while the thick filaments are connected to M-lines. The thick myosin filaments are connected by their myosin head to the thin actin myofilaments. Muscle contraction occurs within sarcomeres by the active movement of the myosin heads along the actin myofilaments.

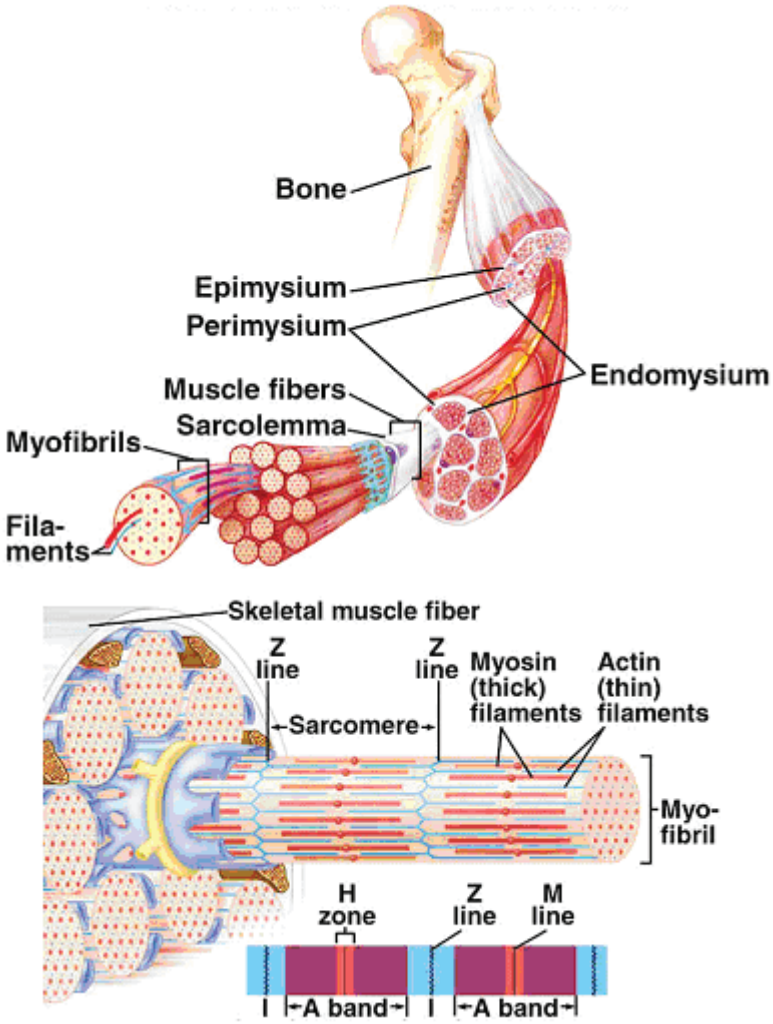

Figure 1. Simplified anatomy of the muscle fibres and myofibrils in muscle (Constantin, 2006).

1.2 Ante mortem muscle physiology and biochemistry

During muscle acitivity the active myosin head can be released from the passive actin myofilaments by utilizing Adenosine Triphosphate (ATP) forming a new myosin to the actin binding site. Active release of calcium ions (Ca^{2+}) in the myofilaments allow the actin binding sites to bind to the myosin heads and to start the muscle contraction process. Resorption of Ca^{2+} by the sarcoplasmic reticulum stops muscle contraction. By repeatedly binding and releasing of actin to myosin, a muscle contraction is generated. When ATP is utilized during muscle contraction of during metabolism, one phosphate is removed from ATP by hydrolysis, which results into Adenosine Diphosphate (ADP).

Available Creatine Phosphate (CrP) in the muscle will transfer its phosphate to the ADP to create a new ATP molecule and act as a direct buffer for generating ATP in myofilaments. When insufficient amounts of ATP are available, the ADP molecules will hydrolyze another phosphate for the release of more energy, forming Adenosine Monophosphate (AMP). This metabolite, AMP, can be broken down into Inosine Monophosphate (IMP) by the enzyme myodenylate deaminase (Nelson, 2005). This occurs when myofilaments have insufficient ATP available for the supply of energy. Elevated IMP levels in the muscle therefore indicate that a severe shortage of muscle energy has occurred.

ATP is continuously generated and metabolised in muscles, and can be generated from carbohydrates, proteins, and is metabolised through aerobic (with oxygen) pathways. The Krebs cycle is the main pathway to produce ATP from carbohydrates in a living pig. Glucose is the required energy source of the Krebs' cycle. The oxidative part of the Krebs' cycle consumes acetate in the form of acetyl-CoA, which is derived from the carbohydrates to generate ATP, carbon dioxide, and water. The total number of ATP generated after complete oxidation of a glucose molecule is estimated between 30 and 38 ATP. The stored muscle glycogen is a glucose source that can be utilized in the Krebs' cycle. The total amount of stored glycogen in myofilaments is called the glycogen potential of a muscle. The glycogen potential can be influenced by many factors such as genetics, feeding strategy, husbandry practices, transport practices, and fasting in live pigs (Sellier & Monin, 1994). Fasting, increased physical activity (i.e. fighting, walking), and longer lasting stress (transport, lairage) will decrease the amount of muscle glycogen at time of slaughter (Bee, 2006; Leheska et al., 2002).

When muscle demands for ATP in the living pig are higher than the amounts generated by the oxidative pathways, the Krebs' cycle can utilize anaerobic (without oxygen) pathways to support muscle cells with additional ATP. The anaerobic energy pathway, or glycolysis, creates ATP exclusively from carbohydrates, with lactic acid as a by-product. Anaerobic pathways generate two lactic acid and two ATP molecules from one glucose molecule and are considered as less efficient than oxidative pathways. During severe shortage of ATP in the muscle, the anaerobic pathway is the only alternative for ATP supply. In a living pig, the generated lactic acid will be transported to other tissues by blood and is metabolised through other tissues via oxidative pathways. Also, blood in the living tissue removes excessive generated heat from tissues.

1.3 Post mortem muscle physiology and biochemistry

After exsanguination, mucle metabolism has to switch from aerobic to exclusively anaerobic pathways for continuing the ATP supply. This will result in accumulation of lactic acid, lowering the muscle pH, and accumulation of heat, due to the absence of blood flow. A low muscle pH in combination with higher muscle temperature will increase occurrence of protein denaturation. The rate and extent of post mortem pH decline therefore runs parallel with reduction of cellular muscle structures (cytoskeletal proteins) by causing protein denaturation and aggregation which will result in pork with a lower water holding capacity. Depending on the glycolitic potential of the muscle, the anaerobic pathways can develop a limited amount of ATP and lactic acid (Van Laack &

Kauffman, 1999). Reduction of muscle glycogen storages in the muscle at slaughter therefore limit the development of post mortem lactic acid accumulation, pH decline, and occurrence of protein denaturation and aggregation.

Acute stress in the animal before slaughter results in elevated levels of stress hormones (cortisol, norepinephrine, and epinephrine) in the muscle. These stress hormones increase the anaerobic muscle metabolism and lactic acid accumulation, and decrease muscle pH in the living animal and muscle early post mortem (Bee, 2006; Briskey & Wismer-Pedersen, 1961; Cannon et al., 1995; Eikelenboom et al., 1974b; Enfalt et al., 1993; Hambrecht, 2004; Hunter et al., 1994).

Minimizing factors which cause acute preslaughter stress for pigs can reduce the speed of post mortem metabolism. Carcass cooling procedures can also influence the post mortem metabolic process, whereby faster chilling will reduce the speed of muscle metabolism. Variation in muscle and fat thickness will directly affect the cooling efficiency, therefore heavily muscled pig carcass will require longer chilling periods before the core muscle temperature is affected, resulting in a faster speed of lactic acid accumulation (Maribo et al., 1998; Offer, 1991; Otto et al., 2004). However, when excessive chilling practices reduce the carcass temperature below 10˚C before the pH is below 6.2, uncontrolled muscle fibre contractions (cold shortening), which toughens the meat can be induced. These findings reveal that there is an optimum cooling procedure (Dransfield & Lockyer, 1985), and that variation on the speed of post mortem metabolism occurs in pork processing plants.

1.4 Post mortem muscle metabolism and pork quality

In general, a high post mortem metabolism results in a low muscle pH early post mortem, which induces occurrence of pork with a lower water holding capacity (WHC). Consumer appreciation and processing yields are affected by pork WHC, and therefore WHC is seen as the most important pork quality parameter. Examples of extremes in pork quality are so-called Pale, Soft, and Exudative (PSE) meat and Dark, Firm, and Dry (DFD) meat. The speed of ATP utilization is normally accompanied by a parallel drop in pH and a gradual reduction in the water binding properties of muscle tissue (Bate-Smith & Bendall, 1947). Pork with (extreme) low pH levels (< 5.4) and severe muscle protein changes, leading to high drip loss% levels, is PSE. Pork with lower glycogen potential at slaughter leading to a higher ultimate pH (>6.2), leading to low drip loss% levels, is DFD. The post mortem muscle metabolism slows down and stops when no more ATP can be formed and ATP depletion leads to the onset of rigor mortis (Greaser, 1986). Pork colour has been related to the WHC, with darker pork having a better water holding capacity (Bredahl et al., 1998; Huff-Lonergan et al., 2002; Mancini & Hunt, 2005).

When post mortem muscle pH decline reaches pH levels lower than 5.6, the negatively charged muscles proteins become closer to their iso-electric point (figure 2). This causes the proteins to denaturate and aggregate, whereby the muscle structure tends to collapse (Bendall, 1962; Cassens et al., 1975; Honikel, 1998; Huff-Lonergan & Lonergan, 2005; Offer, 1991). Protein aggregation and denaturation decreases the capillary forces by enlarging interstitial spaces for the water to escape the structure, which allows water being pushed out when rigor starts (Honikel et al., 1986; Kristensen

& Purslow, 2001; Lawson, 2004; Schäfer et al., 2002). It is known that rigor onset coincides with ATP depletion at generally three through six hours post mortem (Dransfield & Lockyer, 1985; Offer, 1991). Rigor sarcomere shortening of myofibrills enlarges interstitial spaces in muscle for water to escape the structure (Honikel et al., 1986; Lawson, 2004). As a result, less tightly bound water accumulates in muscle and migrates to interstitial spaces (Offer, 1991).

Research shows that the myofibrillar protein system of meat follows not only one simple model of water holding (Puolanne & Halonen, 2010). In pork, water is present intra- and extra-cellular, and can be bound to varying extends (Huff-Lonergan & Lonergan, 2005). Increasing amounts of unbound or less bound water in meat runs parallel with lower water holding capacity and higher drip loss% of meat (Fischer, 2007; Honikel, 1998; Puolanne & Halonen, 2010). Water is held in muscle tissue by chemical bonding, hydrogen bonding, or by capillary forces. Chemically bound water is not released from muscle during the early post mortem period and represents a small proportion of total water in muscle (Huff-Lonergan & Lonergan, 2005). Water held to negatively charged proteins is freed when positively charged ions, like hydrogen ions, accumulate during post mortem muscle metabolism (Bendall, 1962). The speed of post mortem muscle metabolism affects hydrogen and capillary bound water, by reaching the isoelectric point of the muscle proteins (figure 2) whereby muscle proteins reduce water hydrogen bonding, and create interstitial spaces by protein denaturation.

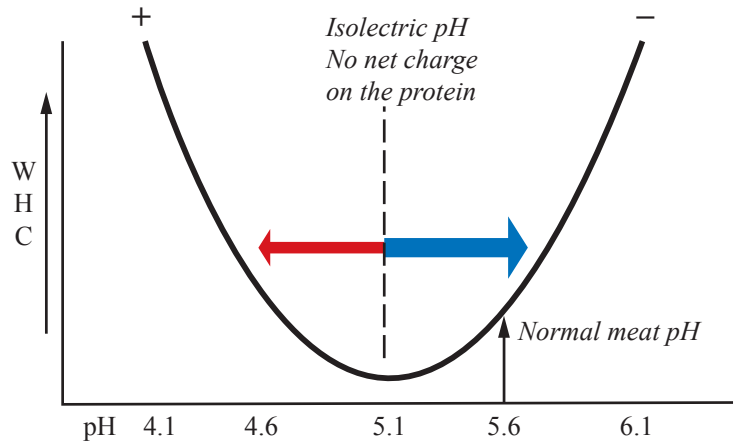

Figure 2. Muscle isoelectric point, pH, and WHC interaction graphically displayed on their importance on pork WHC (Hunt et al., 2011).

During the aging period (> 24 hours post mortem) the muscle cytoskeletal proteins, such as the the Z-disc, M-disc and desmin rich filament structures, are degraded (Kristensen & Purslow, 2001). The degradation process of muscle cytoskeletal protein structures during ageing of meat continue to reduce pork WHC by causing myofibrillar shrinkage of proteins. However, continuous brake down of muscle cytosketelal proteins will create extra-cellular spaces by disconnecting filament structures, which finally reduce post mortem myofibrillar shrinkage and reduce release of water from the meat and increase WHC (Huff-Lonergan & Lonergan, 2005; Kristensen & Purslow, 2001; Offer, 1988b).

1.5 Measuring and prediction of pork water holding capacity

Water accounts for approximately 75% of the muscle weight, and the ability of pork to retain its water is important for both industry and consumers. The content and distribution of water in meat determines its toughness, firmness, juiciness, appearance, and the technological properties of pork during further processing (Bredahl et al., 1998; Cariou et al., 1988; Flores et al., 1999; Puolanne & Halonen, 2010). Intramuscular fat content also correlates positively with juiciness and tenderness of pork, but was shown to be of less significance than WHC (Eikelenboom et al., 1996; Huff-Lonergan & Lonergan, 2005). Pork with low WHC increases cooking losses during further processing (Cariou et al., 1988). An increased water loss during meat preparation prior to consumption results in a less optimal eating quality (Flores et al., 1999). Pork with a relative lower WHC and lower pH value is more favourable for processing of dry cured meat products, such as raw ham and salami. Whereas, for processing of e.g. cooked ham, pork with a relative high WHC is most appropriate. This process optimization in the supply chain can only be realised when there is a tool available to measure and predict pork WHC at slaughter plants (García-Rey et al., 2005; von Rohr, 1999).

Currently, pH, colour L*, and drip loss% measurements are methods to measure and indicate pork WHC. By measuring muscle pH, pork WHC can be predicted, which relates to the rate and extent of post mortem lactic acid accumulation (Eikelenboom et al., 1974b; Kauffman, 1993). Meat colour (L*, a*, and b* values) is also related to pork WHC (Huff-Lonergan et al., 2002; Mancini & Hunt, 2005). There are multiple drip loss% measurements available, which are based on the principal of taking a meat sample of a pre-defined size and expose the sample to gravitation for a specified time period to allow the unbound fluid to exudate from the meat (Honikel, 1998; Offer, 1988b; Otto et al., 2004; Rasmussen & Andersson, 1996). More recently, near infrared spectroscopy (NIRS) was revealed as a potential technique to measure and predict pork WHC (Geesink et al., 2003; Prieto et al., 2009).

1.6 Near infrared spectroscopy (NIRS)

Near infrared spectroscopy is a technology, which measures the reflected light from a (pork) sample that has been exposed to light falling within the near infrared or electro-magnetic region (4000 – 12500 wave number cm^{-1}, Figure 3). Fundamental absorptions and vibrations of light by molecules occur in the infrared region and can be used to reveal information about the components measured (Williams, 2007). Molecules tend to absorb several wavelengths within the NIR region and they often overlap with absorptions of other molecules, which creates complicated absorption spectra (Figure 3). The NIRS technology is capable of measuring multiple wavenumbers ($< 4000\ cm^{-1}$) at the same time and thereby measuring several absorption regions of the sample's molecules (overtones). However, the NIR absorption regions of all molecules from a sample are often complex. The measured absorptions within the reflected light can in some cases be correlated to a chemical or physical parameter, representing the molecule complex in the sample or representing a specific molecule type in the sample. NIRS spectra can be correlated by partial least square regressions (PLS) to the chemical or physical parameter to develop prediction equations, whereby

the corresponding spectral regions are identified and used. In addition spectra data treatments can be applied to enhance correlation of NIRS regions to a specific parameter. The NIRS technique is seen as promising for non invasive inline measurements of carcass parts (Prieto et al., 2009). Studies showed that the coefficient of determination (R^2) of NIRS for drip loss% ranged from 0.20 to 0.71 (Prieto et al., 2009).

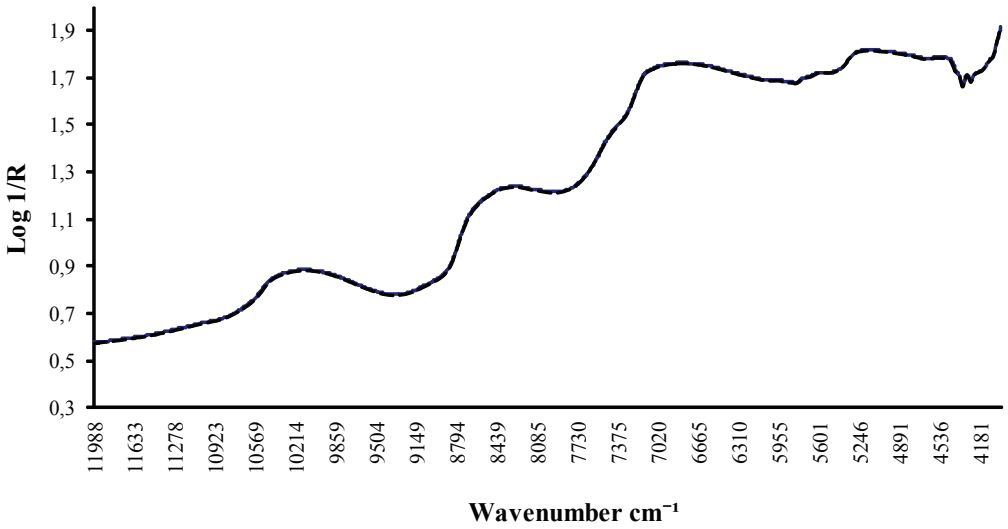

Figure 3. NIRS spectra of pork loin, wavenumber range 4000 – 12000 cm^{-1}, absorption at Log 1/R scale.

2. Aim and outline of this thesis

Water holding capacity of pork defines substantially the sensory appreciation and processing yields. Pork is a natural product and variation occurs. Therefore measuring and prediction of pork WHC is important. The aim of this thesis was to validate rapid prediction of pork WHC. Correlation of fresh meat measurements to pork WHC and its technological quality were investigated. Within this thesis the application of NIRS technology to measure and predict pork water holding capacity and how this relates to the technological quality of further processing pork products was investigated.

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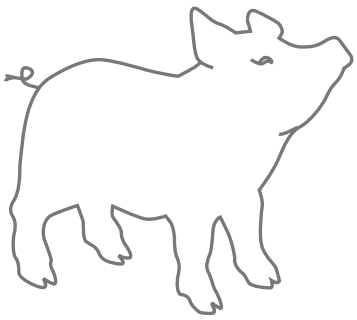

Chapter 1

Prediction of pork quality with near infrared spectroscopy (NIRS)

1. Feasibility and robustness of NIRS measurements at laboratory scale

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Abstract

The objective was to study prediction of pork quality by near infrared spectroscopy (NIRS) technology at laboratory. A total of 131 commercial pork loin samples were measured with NIRS. Predictive equations were developed for drip loss%, colour L*, a*, b*, and pH ultimate (pHu). Equations with $R^2 > 0.70$ and residual prediction deviation (RPD) ≥ 1.9 were considered as applicable to predict pork quality. For drip loss% prediction equation was developed (R^2 0.73, RPD 1.9) and 76% of the grouped superior and inferior samples were predicted within the groups. For colour L*, test-set samples were predicted with R^2 0.75, RPD 2.0, colour a* R^2 0.51, RPD 1.4, colour b* R^2 0.55, RPD 1.5 and pHu R^2 0.36, RPD 1.3. We conclude that NIRS prediction equations could be developed to predict drip loss% and colour L*, of pork samples. NIRS equations for colour a*, b*, and pHu were not applicable for the prediction of pork quality on commercially slaughtered pigs.

Keywords: NIRS, drip loss%, pHu, colour, pork quality

1. Introduction

Drip loss%, colour L*, and pH ultimate (pHu) are important pork quality characteristics and correlate with the sensory appreciation of pork by consumers (Bredahl et al., 1998; Huff-Lonergan et al., 2002). Drip loss% is the amount of intra- and extra cellular fluid separating from the meat. Drip loss%, colour, and pHu also affect the amount of saleable meat by reducing drip loss% and increasing processing yields of further processed products (Bertram et al., 2003). Pork quality is the result of perimortem muscle metabolism, which is influenced by farm management conditions, transport, preslaughter handling, stunning, processing, and cooling conditions of carcasses (Andres et al., 2007; Bowker et al., 1999; Cassens et al., 1975; Channon et al., 2002; Rosenvold & Andersen, 2003). Literature shows that a non-invasive method, such as near infrared spectroscopy (NIRS) is a useful method to measure pork meat quality (Barlocco, 2006; Geesink, 2003; Geesink et al., 2003; Prieto et al., 2009; Rodbotten et al., 2000; Swatland & Barbut, 1995), but a representative sampling method is very important (Brøndum et al., 2000).

A predictive NIRS prediction equation can be evaluated by the coefficient of determination (R^2), standard error of prediction (SEP), and bias. During last decades both NIRS equipment and analyzing software has been used for scientific applications in meat. Some researchers showed promising results with prediction capacities of NIRS for drip loss% with coefficient of determination (R^2) ranging from 0.55 to 0.56 (Brøndum et al., 2000; Savenije et al., 2006). These authors concluded that improved and dedicated methods are needed before these techniques can be introduced on process lines. However, within the pork processing industry there is a need for further market segmentation to fulfil consumer needs with high quality pork products (Barlocco, 2006; Bertram et al., 2003; Brøndum, 1998; Brøndum et al., 2000; Eikelenboom et al., 1974; Enfalt et al., 1993; Fischer, 2007; Forrest et al., 2000; Geesink et al., 2003; Hoving-Bolink et al., 2005; Kauffman, 1993; Leroy et al., 2004; Prevolnik et al., 2005; Rodbotten et al., 2000; Savenije et al., 2006).

The hypothesis was that recent improvements in NIRS technology may have improved the ability to develop prediction equations for pork quality and that reliable prediction equations could be developed. The objective of this study was to investigate the NIRS prediction of pH, drip loss%, and colour L*, a*, b* from intact pork loin samples under laboratory conditions.

2. Materials and methods

2.1 Sample collection and pork quality measurements

A total of 131 *Longissimus dorsi* samples were collected from randomly chosen pigs from a commercial Dutch pork production plant, during two processing days. The pigs were electrically stunned, and had a carcass weight of approximately 92 kg. Samples were taken 24 hours after exsanguination from the *Longissimus dorsi* at the shoulder side at the 4th rib. A rectangular sample of 6 x 5 cm and approximately 2 cm thick was taken. After exposing the sample surface to the air

for more than 30 minutes, colour L*, a*, and b* values (Pulsed xenon lamp, diffuse illumination and silicon photocells detector, Chroma Meter CR-400, Konica Minolta Sensing, Inc.) and pHu (MPI pH-Meter, Meat Probes, Inc) were measured in duplicate. The samples were transported at 4°C to a laboratory in The Netherlands within 1 hour transportation time. A NIRS measurement was taken in the laboratory with a FOSS 6500 (Foss NIRSystems, Silversprings, MD, USA), measuring reflectance from 400 to 2498 nm and 35 scans were averaged per sample. The automatic intake device and the FOSS ¼-rectangular cups were used. The shape of the rectangular samples fitted into the ¼ rectangular cups from the NIRS device. The samples were kept at room temperature (20°C) for approximately 20 minutes before placing in the ¼-rectangular cups to avoid condensation at the quartz glass surface. WinISI III software was used for the analysis of the NIRS measurements. Each NIRS spectrum was automatically transformed into a Log 1/R absorption spectrum by the software.

After the initial NIRS measurements, pork samples were weighed and individually stored at 4°C during 48 hours in consumer retail trays. After 48 hours the samples were weighed again and drip loss% was determined as weight loss percentage of initial sample weight.

The described measurements of drip loss%, colour L*, a*, b*, and pHu were used as the reference parameters for the NIRS data.

2.2 NIRS analysis and data processing

Data were exported from WinISI III software, and the chemometric data treatment was performed using OPUS 6.5 software (Bruker Optics GmbH, Ettlingen, Germany). Spectral repeatability was evaluated using the root mean square (RMS) errors. The measured spectral data were investigated for correlation with the measured reference parameters, using modified partial least square regression (MPLS). Different spectral data pre-treatments were investigated using 1st or 2nd derivative with smoothing, vector normalization, constant offset elimination and no spectral data pre-processing. Also combinations of data pre-treatments were investigated, before spectral data were correlated with the reference parameters. The OPUS 6.5 software offers many options for data pre-processing. The optimal method depends on the dataset studied.

Calibrations were developed for predicting drip loss%, colour L*, a*, b*, and pHu. Evaluation of reference data is important, since the calibration is dependent on the quality of the reference data. Therefore the reference data were evaluated by the mean, standard error (SE), range, coefficient of determination (R^2), and standard error of laboratory (SEL). The SEL was defined as the standard error of variance between duplicates analysed by the reference method. The SEL was calculated by using the following equation:

$$SEL = \sqrt{\frac{\sum_{i=1}^{n}(y_1 - y_2)}{2n}}$$

Where y1 – y2 is the difference between duplicate measurements by the reference method on sample i. For pHu and the colour L*, a*, and b* values replicates were measured and SEL was calculated.

Prediction equations were created using OPUS 6.5 software with using MPLS. The residuals of each wavelength were obtained after each factor and were individually calculated. These residuals were standardised in MPLS (divided by the standard deviations of the residuals at a wavelength) before calculation of the next factor (Flores et al., 2009). The number of factors in the chemometric prediction equation is shown as the "rank". The chemometric prediction equation was set to a maximum rank of 10.

During the development of prediction equations, outliers were removed following the next equation;

$$Outlier = \mathrm{A} - \left(\overline{\mathrm{A}} \pm 2 * SD\right) \quad ,$$

whereby A was the difference between the actual and predicted value of a reference parameter.

The difference between the actual and predicted reference values during validation of the prediction equation, using independent test-set samples, was calculated.

2.3 Calibration and test-set

Before developing prediction equations the data set was split into two datasets; a calibration-set (92 samples) and a test-set (39 samples). The test-set samples were selectively chosen by the software, by which samples were equally distributed over the spectra variation to cover the whole variation range in both datasets.

The calibration-set was used to develop prediction equations, using the MPLS, to predict the measured meat quality parameters. The calibration-set was evaluated by using cross-validation. For cross-validation every single spectral sample is validated using a calibration prediction equation developed on the other samples of that data-set. The validation errors of those individual samples are combined in a root mean square error of estimate (RMSEE). The prediction equations developed from the calibration-set were also evaluated on the independent test-set samples, by using test-set validation. For test-set validation every spectral sample was taken out and validated using the prediction equation developed on the calibration samples. The validation errors from the test-set are combined in a root mean square error of prediction (RMSEP).

The spectra following data pre-treatments were investigated, using 1st or 2nd derivative with smoothing, vector normalization, constant offset elimination and no spectral data pre-processing. Also combinations of data pre-treatments were investigated, before spectral data were correlated with the reference parameters.

No signal noise at the beginning and the end of the spectral range was eliminated. The whole spectral region at 2 nm intervals, was selected for performing the calibration prediction equations; 408-2492 nm, including the visual regions from 408-1100 nm. The performance and reliableness

of the prediction equations were evaluated using the following statistics: the root mean square error of estimate (RMSEE), rank, root mean square error of prediction (RMSEP), residual prediction deviation (RPD), and the coefficient of determination (R^2). The RPD shows the ratio between the standard deviation (SD) of the original reference data to the root mean square error of estimation (RMSEE) or the root meat square error of prediction (RMSEP). The RPD was calculated using the following equations:

$$RPD = \frac{SD_{ref}}{RMSEE(or)RMSEP}$$

Prediction equations with $R^2 \geq 0.70$ and $RPD \geq 1.9$ were considered as applicable to predict pork quality.

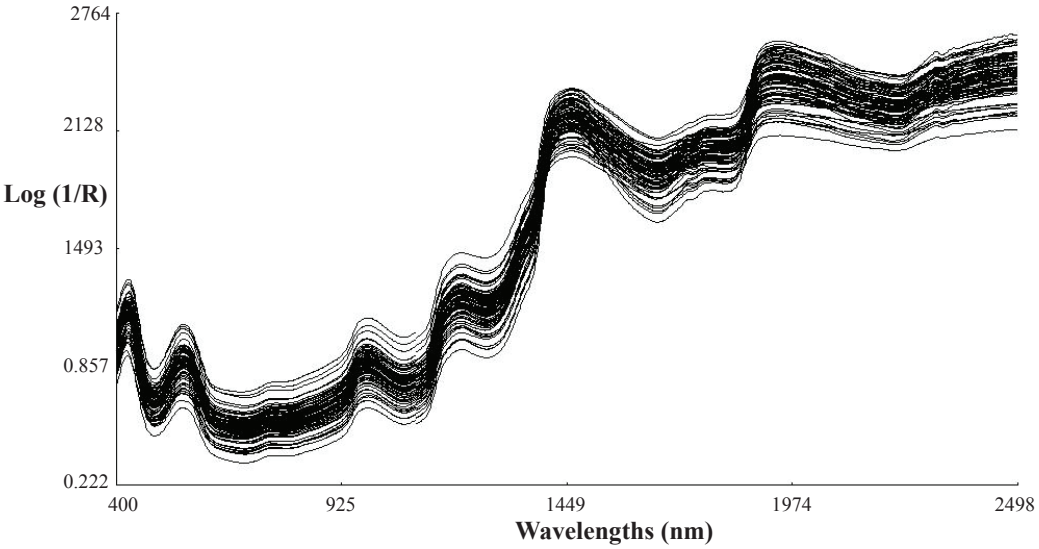

Figure 1. NIRS spectra of all fresh pork samples measured 24 hour post mortem.

3. Results and discussion

3.1 Spectra collection and pre-processing

The quality of the spectral and reference data is critical for the subsequent development of prediction equations for estimation of the various meat quality parameters. The spectra displayed low noise levels, and the spectral absorption regions ranged between 0.5 and 2.7 Log 1/R (Figure 1, WinISI III software).

Table 1. Mean, standard deviation (SD), range, coefficient of variation (CV), and standard error of laboratory (SEL) of the reference pork quality parameters.

Parameter	Mean	SD	Range			CV	SEL
L*	56.1	4.2	47.3	-	66.4	7.5	1.2
a*	8.6	1.6	5.1	-	13.3	18.6	0.6
b*	7.9	1.8	3.6	-	12.1	23.1	0.6
pHu	5.7	0.2	5.3	-	6.4	3.8	0.1
Drip loss%	3.0	1.4	0.7	-	7.0	46.1	n.a.

Some researchers showed that the composition of ground meat samples can be more accurately predicted than intact meat samples with NIRS (Barlocco et al., 2006; Prevolnik et al., 2005; Prieto et al., 2009). Intact muscle fibres may act as optical fibres and tend to conduct light along their length and by internal reflections. Intact muscle fibres tend to absorb more energy in comparison with minced samples (Prieto et al., 2009). However, it is more likely that online application of NIRS will be performed with non-destructive sample measurements. It is proven that by choosing the optimal number of scans and scanning area the ability of NIRS to predict the composition of intact meat samples can be improved (Prevolnik et al., 2005; Prieto et al., 2009; Rodbotten et al., 2000).

Figure 2 shows the full spectrum data modified to the 1st derivative, gap of 5 datapoints, smoothing over 5 datapoints and final smoothing over 1 data points (1,5,5,1), performed with WinISI III software. With pre-treatment of NIRS spectral data, systematic variations, and baseline variations unrelated to the reference parameters can be reduced. Therefore, caution is needed since pre-treatments can also remove significant information from the spectra (Azzouz et al., 2003). The partial least square regression (PLSR or PLS) method compares multiple wavelengths with reference parameters.

3.2 Calibration and prediction

Table 1 shows the mean, standard deviation (SD), range, coefficient of variation (CV), and standard error of laboratory (SEL) of the reference pork quality characteristics. The SEL for drip

loss% could not be calculated since no repetitions were taken. The relative SD was 19 – 22% of the difference between the maximum and minimum value of the reference parameters. Therefore this dataset had a sufficient variation for the development of meaningful prediction equations. Correlation coefficients squared (r^2) of the reference parameters are given in table 2. During the development of a prediction equation for drip loss% and pHu, 2 samples were considered as outliers and removed from the calibration data set. One sample from the drip loss% test-set validation was considered an outlier and was removed. The correlation coefficients (r) to the reference parameters range from – 0.20 up to 0.70, and are in similar range, compared with data of prior studies (Forrest et al., 2000; Geesink, 2003; Geesink et al., 2003; Pedersen et al., 2003; Prieto et al., 2009).

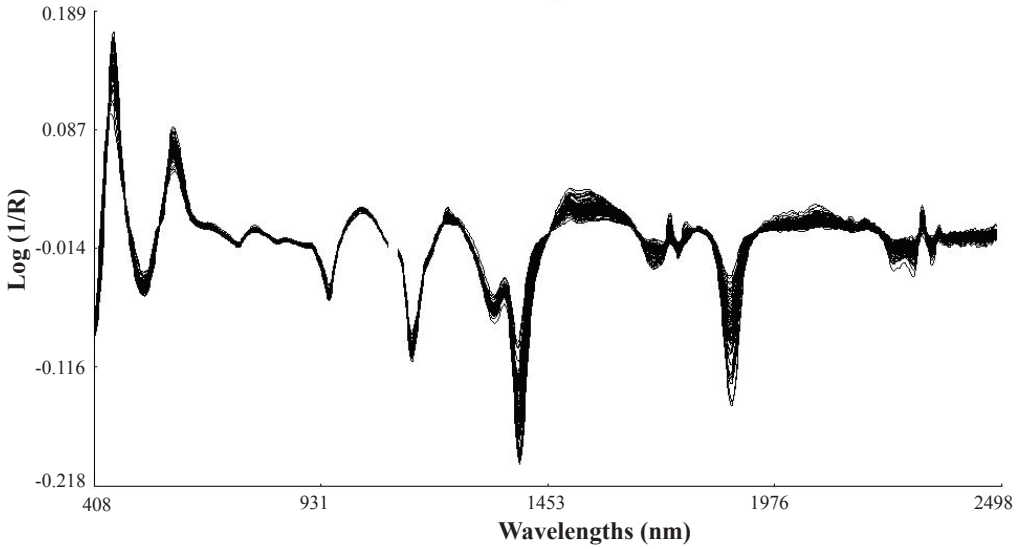

Figure 2. NIRS spectral data modified to the 1st derivative, gap of 5 datapoints, smoothing over 5 datapoints and final smoothing over 1 datapoint (1,5,5,1), performed with WinISI III software. X-axis expresses the wavelength in nanometers of the spectra and the Y-axis expresses the absorption values (Log 1/R) of the modified datapoints.

Table 2. Correlation coefficient matrix (r^2) of measured reference pork quality parameters.

	Colour a*	Colour b*	pHu	Drip loss%
Colour L*	0.10	0.60	0.04	0.43
Colour a*	-	0.50	0.00	0.03
Colour b*	-	-	0.01	0.20
pHu	-	-	-	0.09

Table 3. Calibration statistics obtained for the prediction of Colour L*, a*, b*, pHu and drip loss% in commercially slaughtered pigs, by using an independent test-set validation dataset of 39 samples.

	Region 1	Region 2	Region 3			Calibration						Test-set validation				
Parameter	(nm)	(nm)	(nm)	Treatment	Smooth	Mean	n	R^2	RMSEE	RPD	Rank	n	R^2	RMSEP	RPD	Bias
L*	800-1336	1638-1837	2172-2261	A	25	56.1	92	0.76	2.1	2.0	8	39	0.74	2.3	2.0	0.1
a*	800-1336	None	none	A + C	25	8.6	92	0.54	1.1	1.5	7	39	0.51	1.2	1.4	0.1
b*	1332-1644	1830-2182	none	D	none	7.9	92	0.48	1.3	1.4	7	39	0.55	1.3	1.5	0.1
pHu	436-481	806-972		B	25	5.7	92	0.39	0.2	1.3	4	39	0.36	0.2	1.3	0.1
Drip loss%	800-1336	1638-2182	none	A	13	3.0	90	0.80	0.6	2.2	7	38	0.73	0.8	1.9	0.0

Regions are expressed in nanometres. Treatment A is 1st derivative, B is 2nd derivative, C is vector normalization and D is constant offset elimination.

Characteristics of the NIRS prediction equations are given in table 3. The root mean square error of estimation (RMSEE) or the root mean square error of cross validation (RMSECV) indicates the error of the calibration dataset. The root mean square error of prediction (RMSEP) is the error of the test-set. In our dataset, the difference between the RMSEE and the RMSEP of the test-set varied between 0.1 and 0.2. This relatively small difference indicated that the prediction equation developed from the calibration set gave similar results when used on independent samples of the test-set. The difference between the RMSEP and the standard error of laboratory (SEL) ranged from 0.1 to 1.0 and was 0.6 on average. The RMSEP showed higher errors than the SEL, which was expected since the prediction equations are developed from the reference values, and therefore include the SEL of these reference values. The chosen prediction equation for drip loss% showed an RMSEP of 0.79 and an RMSEE of 0.61 which is lower than the RMSEP reported by Geesink et al., (2003) [1.1] and Forrest et al., (2000) [2.43]. The RMSEE from the drip loss% prediction equation is also considerably lower than the RMSEE reported by Pedersen et al., (2002) [0.85%] and Savenije et al., (2006) [1.24%]. The observed lower RMSEE and RMSEP from our data indicate an improved prediction equation, based on improved data pre-treatments, different drip loss% method, or less systematic and unrelated variations in the spectral data, in comparison to the other studies.

Within our dataset, the samples could be grouped on an arbitrary basis. Samples with < 2.0% drip loss% could be ranked as having superior water holding capacity, and samples > 4.0% drip loss% as having inferior water holding capacity. In this study 32% (n = 12) of the test-set samples were classified as superior (drip loss% < 2%) and 24% (n = 9) were classified as inferior (>4%). However the drip loss% as predicted from the prediction equation resulted in 21.1% (n = 8) as superior and 26.3% (n = 10) as inferior. The predicted values and the observed drip loss% on the test-set samples are shown in figure 3. From the superior group 3 samples were predicted as having more drip loss% than 2% (e.g. 2.06%, 2.11%, 2.13%) while from the inferior group 2 samples were predicted as less than 4% drip loss% (e.g. 2.94% and 3.91%). From the superior group 3 out of 12 samples were categorized incorrectly (25%) and from the inferior group 2 out of 9 samples (22%) were categorized incorrectly. None of the incorrect superior samples where categorized to the inferior group and vice versa. Savenije et al., (2006) used grouping limits of <6% as superior and >8% as inferior and showed that 50% within the superior group was categorized incorrectly while within the inferior group 36% was categorized incorrectly. It should be noted that Savenije et al., (2006) used 8 circular (thickness 1.8 cm and diameter 4.0 cm) samples per animal and their samples were placed on display trays and covered with PVC cling film and stored for 5 days at 4.0°C. Their drip loss% method resulted in higher drip loss% values and therefore different thresholds in comparison with our study.

In the present work, data pre-treatments were used for the development of all prediction equations. The used amount of factors (rank) for the prediction equations varied between 4 and 8. The rank is the amount of factors which is needed to explain the reference parameters from the spectral data. Generally a lower rank is preferred over a high rank (>10) to avoid over fitting of the prediction equation (Stchur et al., 2002). The RPD value can be seen as an indicator of the ratio between the standard deviation of the reference values and the error of prediction (RMSEE or RMSEP). Generally a RPD > 3 is considered as a reliable prediction equation, while 2 > RPD < 3 can

be seen as a promising prediction equation, which still needs improvement. In theory the RMSEP should ≥ RMSEE, since the developed prediction equation is tested on independent samples. The chemometric prediction equations show higher RMSEP than their RMSEE values. The bias shown on the test-set dataset indicates that the chemometric prediction equation shows similar slopes for both the test-set dataset and the calibration dataset. Despite the fact that the correlations are not very high and the RPD are ≈ 2.0, NIRS could be used for screening of colour L* and drip loss% ($R^2 > 0.70$ and RPD > 1.9). The results are based on samples from commercially slaughtered and processed pork, and can be considered as promising for the use of selecting and sorting for meat quality (drip loss% and colour L*) at slaughterhouses. Figure 3 and 4 show the relation between the observed values and predicted values for drip loss% and colour L*. In comparison with studies of others our prediction equations for drip loss% and colour L* are promising (Barlocco, 2006; Brøndum et al., 2000; García-Rey et al., 2005; Geesink et al., 2003; Hoving-Bolink et al., 2005; Leroy et al., 2004; Prieto et al., 2009; Rodbotten et al., 2000; Savenije et al., 2006). Thus, Williams (2007) stated that an $R^2 > 0.70$ and RPD ≥ 2.0 are minimal requirements for a prediction equation and can only be used for rough screening, and need substantial improvement (Williams, 2007).

It should be stated that rapid screening of meat quality is of great interest for the meat industry and therefore non invasive screening could be promising for a practical application (Barlocco, 2006; Bertram et al., 2003; Brøndum, 1998; Brøndum et al., 2000; Eikelenboom et al., 1974; Enfalt et al., 1993; Fischer, 2007; Forrest et al., 2000; Geesink et al., 2003; Hoving-Bolink et al., 2005; Kauffman, 1993; Leroy et al., 2004; Prevolnik et al., 2005; Rodbotten et al., 2000; Savenije et al., 2006). The R^2 and RPD for colour a*, b*, and pHu showed a coefficient of determination (R^2) < 0.70 and RPD < 2.0, which indicates that the prediction equations are less reliable and unlikely to be used for sorting with NIRS on commercially slaughtered pigs. Practical applications generally require an R^2 of higher than 0.70 and a RPD ≥ 2.0 (Prieto et al., 2009; Williams, 2007; Williams & Sobering, 1993). To reduce the effects of the variation of pork and improve the prediction equations Hoving-Bolink et al., (2005) stated that a large scanning area is necessary. Scanning a larger area by applying more scans will result an increase in the minimal scanning time needed per sample. Increasing the scanning area could have improved our prediction equations. However, increasing the scanning area might extent the scanning time which is a critical limitation for a practical application. Despite the potential of NIRS to predict some technological and sensory attributes on pork it is still unknown if NIRS can be used for online applications on pork in processing plants.

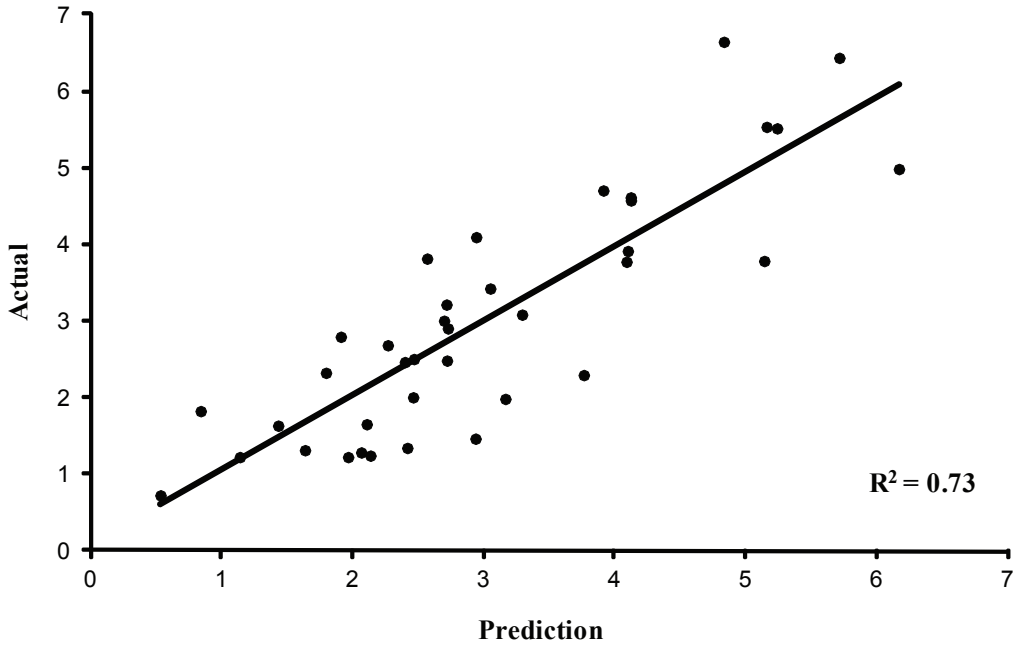

Figure 3. Predicted and actual measured drip loss% on validation samples (test-set), using the prediction equation from the calibration-set.

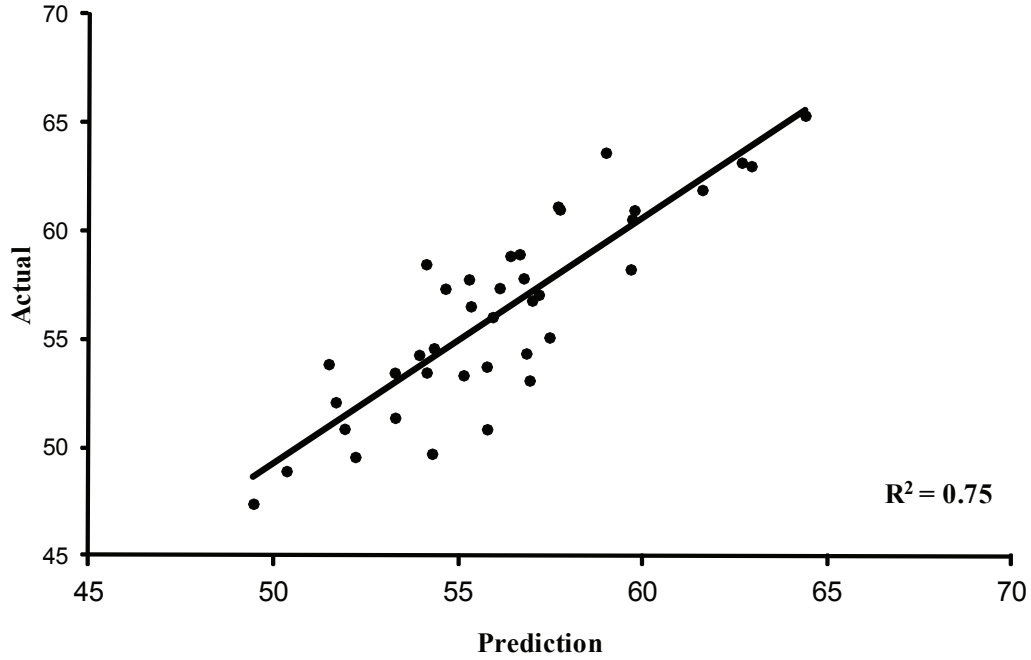

Figure 4. Predicted and actual measured colour L* on test-set samples, using the prediction equation from the calibration-set.

4. Conclusions

The developed prediction equations for drip loss% and colour L* showed a higher R^2 and lower RMSEE and RMSEP in comparison with some other studies. The RPD of the prediction equations drip loss% (RPD= 1.9) and colour L* (RPD= 2.0) did meet the minimal requirements of 1.9. However, improvements should be made. Colour a*, b*, and pHu resulted in an $R^2 < 0.70$ and a RPD < 1.9 and did therefore not generate reliable prediction equations. The prediction equations for drip loss% and colour L* were developed with ranking lower than 10 and indicate that most information about the parameters could be derived within the first factors and over fitting did not occur. The prediction equations for drip loss% and colour L* have the potential to predict some of the main pork quality aspects on intact meat and classify meat into quality groups.

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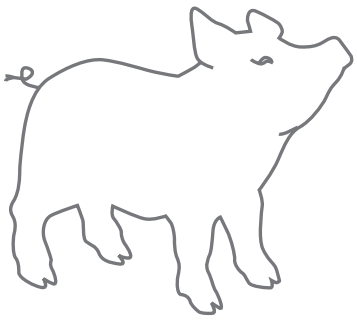

Chapter 2

Prediction of pork quality with near infrared spectroscopy (NIRS)

2. Feasibility and robustness of NIRS measurements under production plant conditions

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Abstract

Longissimus dorsi samples (685) were collected at four processing plants and were used to develop prediction equations for meat quality with near infrared spectroscopy. Equations with $R^2 > 0.70$ and residual prediction deviation (RPD) ≥ 2.0 were considered as applicable for screening. One production plant showed R^2 0.76 and RPD 2.05, other plants showed $R^2 < 0.70$ and RPD < 2.0 for drip loss%. RPD values were ≤ 2.05 for drip loss%, for colour L* $\leq$ 1.82, and pH ultimate (pHu) ≤ 1.57. Samples were grouped for drip loss%; superior (< 2.0%), moderate (2-4%), inferior (> 4.0%). 64% from the superior group and 56% from inferior group were predicted correctly. One equation could be used for screening drip loss%. Best prediction equation for colour L* did not meet the requirements (R^2 0.70 and RPD 1.82). pHu equation could not be used. Results suggest that prediction equations can be used for screening drip loss%.

Keywords: NIRS, drip loss%, pHu, colour, pork quality

1. Introduction

Pork quality is the result of perimortem muscle metabolism, which is influenced by live animal characteristics, pre-slaughter handling conditions, stunning and culling procedures and carcass cooling (Andres et al., 2007; Bowker et al., 1999; Cassens et al., 1975; Channon et al., 2002; Rosenvold & Andersen, 2003). During the last 10 years there has been a focus on how the quality of pork influences the consumer sensory appreciation of pork (Bredahl et al., 1998; Huff-Lonergan et al., 2002). Pork quality also reduces the amount of saleable meat by reducing drip loss% and by increasing processing yields of further processed products (Bertram et al., 2003). The pork processing industry could further segment the market to fulfil consumer needs with high quality pork products (Barlocco et al., 2006; Bertram et al., 2003; Brøndum et al., 1998; Brøndum et al., 2000; Eikelenboom et al., 1974; Enfalt et al., 1993; Fischer, 2007; Forrest et al., 2000; Geesink et al., 2003; Hoving-Bolink et al., 2005; Kauffman, 1993; Leroy et al., 2004; Prevolnik et al., 2005; Rodbotten et al., 2000; Savenije et al., 2006). A potential non-invasive method like near infrared spectroscopy (NIRS) could be useful to measure pork quality (Barlocco et al., 2006; Kapper et al., 2012; Swatland & Irie, 1992).

Earlier studies showed that the coefficient of determination (R^2) of prediction equations by NIRS for i.e. drip loss% ranged from 0.55 to 0.73 (Brøndum et al., 2000; Kapper et al., 2012; Savenije et al., 2006). Rapid and non invasive screening with NIRS could be useful as a practical application for sorting for pork quality (Kapper et al., 2012). However, studies on NIRS spectra measured under cold room conditions (~ 4°C) have not been intensively investigated on pork with using NIRS Fourier-transform (FT) technique and a contact free probe.

The objective of this study was to investigate the prediction of pHu, drip loss%, and colour of NIRS data, measured on pork loin samples, captured under cold room conditions.

2. Materials and methods

2.1 Sample collection and pork quality measurements

A total of 685 *Longissimus dorsi* samples were collected from randomly chosen pigs at four Dutch pork processing plants (A, B, C, and D) during one processing day at each plant. The number of samples varied per production plant (158 – 192). The pigs were electrically stunned with a carcass weight of approximately 92 kg. Samples were taken 24 hours after exsanguination. The sampling procedure was followed as described by Kapper et al., (2012) and samples were taken at 24 hours after exsanguination from the *Longissimus dorsi* at the shoulder side at the 4th rib. A slice of approximately two cm thick was taken from the *Longissimus dorsi* for the determination of drip loss% (48 hours). After exposing the sample surface to the air for more than 30 minutes, colour L*, a*, and b* values (Pulsed xenon lamp, diffuse illumination and silicon photocells detector, Chroma Meter CR-400, Konica Minolta Sensing, Inc.) and pHu (MPI pH-Meter, Meat Probes, Inc) were

measured of the *Longissimus dorsi*. A NIRS measurement on the fresh cut surface was directly taken after the slice (for drip loss%) was sampled in the cold room (~ 4°C) of the production plants. Spectra were measured with a Bruker Matrix-FE (Bruker Optics GmbH, Ettlingen, Germany) using a contact free probe (Q-412 Bruker Optics GmbH, Ettlingen, Germany) with an integrated light source. The device measured reflectance from 4.000 to 12.000 cm^{-1} and 32 scans were averaged per sample. The chosen wavelength resolution was 16 cm^{-1}. The NIRS settings were chosen to be close to the maximal scanning time for practical application. The total scan time was 10 seconds per sample. The NIR spectrum was automatically transformed into Log 1/R absorption spectrum by the software and saved for further analysis.

After the NIRS spectra were taken, the pork slices were weighed and individually stored at 4°C during 48 hours in consumer retail trays. The meat slices were stored at the production plant where the measurements were performed. Colour L*, pHu, and drip loss% were used as reference parameters for the NIRS data. The temperature of carcasses and the air temperature of the chilling rooms were measured for the characterisation of the cooling performance of the four slaughter plants. The used temperature loggers were set at a sampling interval of every two minutes and the insertion tip of the probe was inserted in the middle of the *Longissimus dorsi* (Temprecord International Ltd, Greenmount, Auckland, New Zealand, Multitrip ™). Protocols were followed as described by (Kurt & Klont, 2007). The pH was measured on every single carcass at 35 minutes, 3 hours, 6 hours and 24 hours after exsanguination. Air temperature was recorded with three temperature loggers and the core temperature of the *Longissimus dorsi* was recorded with six temperature loggers, in six different carcasses. The air and carcass temperature data were averaged to generate an overview of the cooling capacity of a production plant. The pH curves could be used to identify the rate of lactic acid accumulation as a result of the speed of post mortem muscle biochemistry.

2.2 Analysis and data processing

NIRS spectra were recorded with a Matrix-F and a contact free probe (Q412) with external light source (Bruker Optics GmbH, Ettlingen, Germany). The device used Fourier transformation and captured reflected light during backward and forward movements of the mirrors (2- sided) to allow short scanning time. Samples were presented once to the probe. During the sample presentation, 32 scans were taken per sample with a resolution of 16 cm^{-1}. Chemometric data treatments were performed with OPUS 6.5 software (Bruker Optics GmbH, Ettlingen, Germany). Reference data were evaluated by the mean, standard error (SE), range, correlation coefficient, and the coefficient of variation (CV %).

Table 1. Mean, standard deviation (SD), range and coefficient of variation (CV%) of the reference pork quality parameters grouped by location and combined together.

Location		mean	SD	min	max	CV(%)
A	Colour L*	54.2	5.5	39.1	65.1	10.2
B		54.1	4.4	42.7	67.2	8.1
C		57.4	5.1	42.7	67.2	8.9
D		55.8	4.1	45.0	63.8	7.3
All		55.3	5.0	39.1	67.2	9.0
A	pHu (Log)	5.69	0.17	5.23	6.42	3.0
B		5.60	0.15	5.30	6.20	2.7
C		5.65	0.13	5.42	6.17	2.3
D		5.58	0.12	5.21	5.95	2.1
All		5.63	0.15	5.21	6.42	2.7
A	Drip loss% (48h)	2.1	1.7	0.5	9.8	79.7
B		1.7	1.2	0.5	6.7	69.2
C		2.7	1.5	0.7	6.8	53.7
D		3.0	1.8	0.6	7.8	61.4
All		2.3	1.6	0.5	9.8	69.3

2.3 Calibration and test-set

Before developing prediction equations, five datasets were created. The data were grouped by production plant (A, B, C, D) and all data were combined in one dataset. Principal component analysis was performed on the spectral data to identify possible spectral outliers and to identify spectra differences between production plants. All created datasets were split into two datasets; a calibration-set (50% of the samples) and a test-set (50% of the samples). The test-set samples were selectively chosen by the software to get an equal distribution of the spectra variation in both datasets.

The calibration-set was used to develop prediction equations. The chemometric prediction equation was set to a maximum rank, or factors, of 10. The calibration-set was evaluated by using cross-validation. Samples with a high Mahalanobis distance versus spectral residual (>2.5) were regarded as less likely to fit to the dataset and were removed, during calibration. Whole spectral region of 4.000 to 12.000 cm^{-1} with 16 cm^{-1} resolution, was used for developing the calibration prediction equations. Spectral regions within the whole region and combinations of regions were investigated for the best informative areas for developing prediction equations. The best performing

regions were investigated for its correlation with the measured reference parameters, using modified partial least squares regressions (MPLS). General variation from environmental factors during scanning (such as sample presentation) could be reduced by using spectral data pre-treatments (Azzouz et al., 2003). The contribution of spectra pre-treatments were investigated during the development of prediction equations. Different pre-treatments were investigated using: no spectral data pre-processing, constant offset elimination, straight line subtraction, vector normalization, min-max normalization, multiplicative scatter correction, 1st or 2nd derivative with smoothing and 1st derivative with multiplicative scatter correction. Also combinations of data pre-treatments were investigated, before the optimal prediction equation was selected.

The optimal data pre-treatment and the optimal spectral regions were selected, based on the cross-validation results and can be found in table 3. The selection criterion for the optimal pre-treatment and regions were a combination of the highest coefficient of determination (R^2), lowest root mean square error of estimation (RMSEE), the highest residual prediction deviation (RPD), and limitation of the number of factors (≤ 10). The RPD value was used as an indicator of the ratio between the standard deviation of the reference values and the error of prediction.

3. Results and discussion

3.1 Reference data

Table 1 shows the mean, standard deviation (SD), range of the minimum and maximum value and CV% for colour L*, pHu and drip loss% at every location. A high CV% indicates high variation in the dataset which is more favourable for developing prediction equations. The observed CV% values indicate that drip loss% had substantial variation that generally is needed to develop prediction equations. The dataset of plant A showed the highest CV% (79.9 %) for drip loss%. Correlation coefficients (r^2) of the reference parameters are given in table 2. The overall results of the pH development and cooling curves are shown in table 3 and figure 4, 5, 6, and 7 and indicate that the cooling was different between the plants, which might explain the differences in mean and standard deviation on meat quality parameters between those plants. The cooling curve of plant A was moderate, when compared with the other plants. The early post mortem measurements of pH 45 minutes, pH 3 hours and pH 6 hours in plant A showed high variation in comparison with other plants, which could explain the higher amount of variation in meat quality at plant A. Despite the same type of stunning devices were used in the four plants, there might be differences between the performance and output of the stunning devices which might influence the early post mortem muscle metabolism and therefore the drip loss%.

Table 2. Correlation coefficient matrix (r^2) of measured reference pork quality parameters.

Location		Drip loss%	L*
All	L*	0.47	
	pHu	0.17	0.15
A	L*	0.47	
	pHu	0.22	0.25
B	L*	0.32	
	pHu	0.10	0.02
C	L*	0.45	
	pHu	0.19	0.30
D	L*	0.68	
	pHu	0.28	0.24

3.2 Spectra and pre-processing

The obtained spectral data did visually display low noise levels. Main noise levels were found in the spectral region between 4000 and 4500 cm^{-1}, which is shown in figure 1. The spectral absorption region of 4000 till 5200 cm^{-1} ranged between 1.0 and 2.8 Log 1/R. An absorption level of 2.8 Log1/R means that only 0.16% of all emitted light reached the detector, which can be considered as a very limited amount of useful data. It is known that intact muscle fibres or myofibrils may act as optical fibres and tend to conduct light along their fibrils and by internal reflections (Prieto et al., 2009). The conduction of light along the muscle fibres might have contributed to the relatively high absorption levels of the spectra at the region 4000 till 5200 cm^{-1}.

Figure 2 shows that evaluation with principal component analysis at factor 2, spectral data of all production locations was evenly distributed and no spectral differences between production locations were observed. This indicates that the data could be combined in one dataset. Generally, effects generated by sample preparation, such as baseline differences, can be reduced by using spectral pre-treatments. For this reason it is more favourable to apply a pre-treatment on spectra, such as first derivative rather than not using any pre-treatment. However, the developments of the prediction equations for drip loss% showed that the best results were gained using no spectral pre-processing.

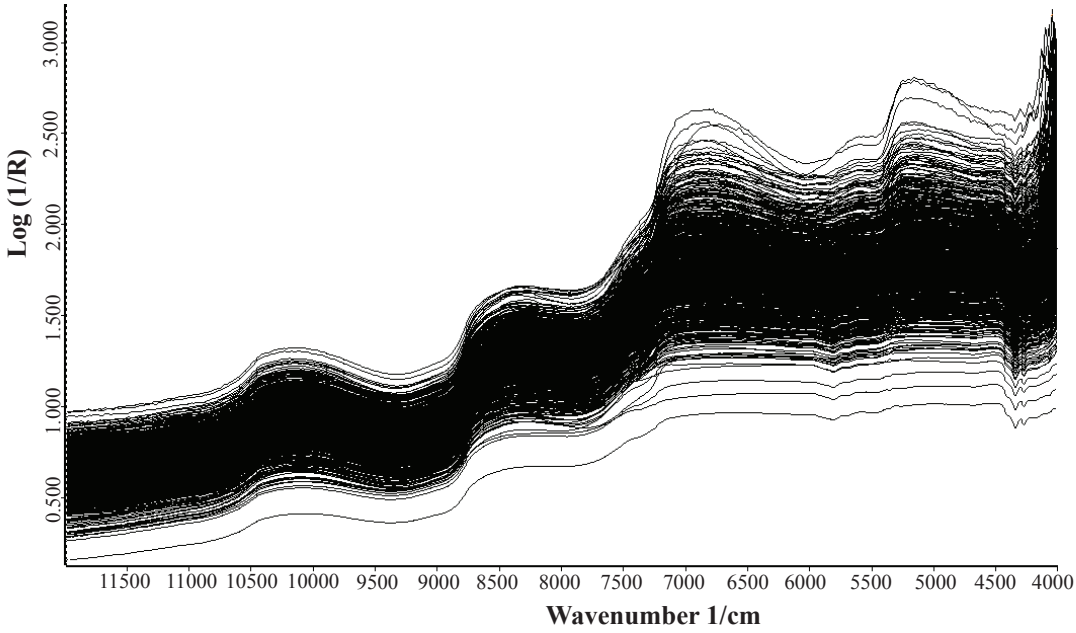

Figure 1. NIRS spectra of all fresh pork samples measured 24 hour post mortem.

3.3 Calibration and test-set results

During the development of prediction equations a total of 685 samples were used. The coefficient of determination (R^2) for the most important reference parameter, drip loss%, ranged from 0.54 till 0.76 on the test-sets between different productions locations. Comparable results were found in other studies (Forrest et al., 2000; Geesink et al., 2003; Pedersen et al., 2003; Prieto et al., 2009). Characteristics of the NIRS prediction equations are given in table 4. In our datasets, the difference between the RMSEE and the RMSEP for drip loss% was < 0.1. This relatively small difference and the relatively small difference between the R^2 values of the equations indicated that the developed prediction equations for drip loss% are likely to be consistent when they would be used on other samples. The chosen prediction equations for drip loss% used the following spectral regions: 11965 – 10368, 8779 – 7977 and 6388 – 4791 cm^{-1}. For colour L*, regions of 11170 – 10368, 9581 – 7977 and 6388 – 5585 cm^{-1} were used. For pHu regions 11965 – 9574, 8779 – 7182 and 5593 – 3996 cm^{-1} were used. The chosen prediction equations for drip loss% showed a RMSEP of 0.73 – 1.12, which is comparable with Geesink et al. (2003) (1.1) and lower than Forrest et al. (2000) (2.43). The RMSEE from the drip loss% prediction equation (0.80 – 1.11) is comparable with Pedersen et al. (2002) (0.85) and Savenije et al. (2006) (1.24). For colour L*, the RMSEP varied (2.82 – 3.6) and was higher than the RMSEP found by Savenije et al. (2006) (1.25- 1.64).

Table 3. pH values measured on the individual carcasses at 45 minutes, 3 hour, 6 hour and 24 hours after exsanguination.

Location	pH 45 minutes (SD)	pH 3 hour (SD)	pH 6 hour (SD)	pH 24 hour (SD)
A	6.17 (0.25)	5.98 (0.22)	5.78 (0.17)	5.69 (0.17)
B	6.49 (0.18)	6.21 (0.23)	5.99 (0.19)	5.60 (0.15)
C	6.38 (0.25)	6.08 (0.25)	5.73 (0.19)	5.64 (0.13)
D	6.39 (0.21)	6.08 (0.25)	5.81 (0.17)	5.58 (0.12)
All	6.36 (0.25)	6.09 (0.25)	5.83 (0.20)	5.63 (0.15)

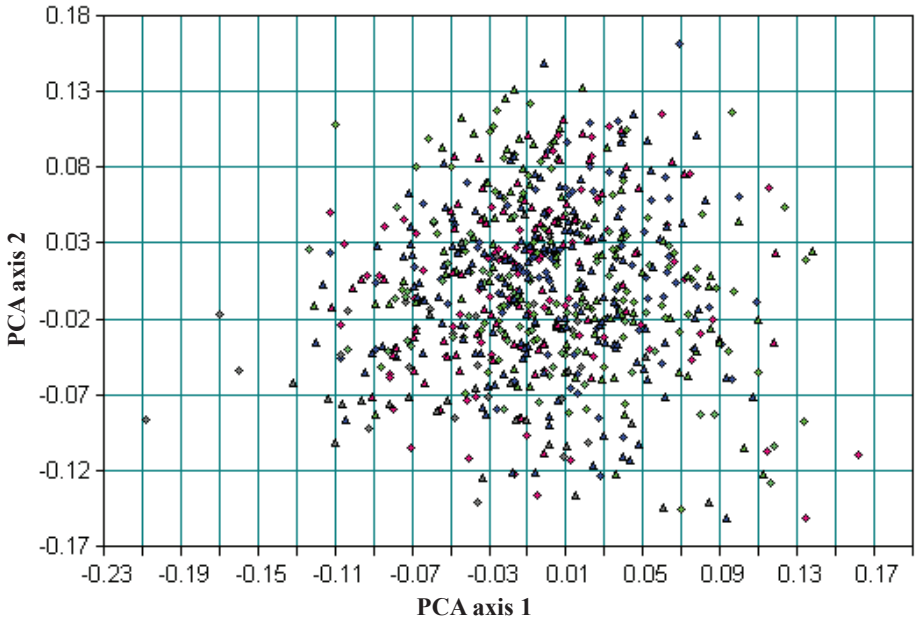

Figure 2. Principal component analysis at factor two of NIRS spectra from 4 different production plants, to identify spectral differences between production plant data.

3.4 Screening for drip loss% with NIRS

Within the 335 test-set samples of the combined dataset, samples could be grouped on an arbitrary base into three classes. Samples with value < 2.0% drip loss were ranked as having superior water holding capacity and samples with value > 4.0% drip loss as having inferior water holding capacity. On the test-set samples 55% were found superior (n = 185), while 16% (n = 52) were found inferior. The grouped samples could be used for the validation of the developed NIRS prediction equation from the calibration samples. The developed prediction equation from the combined dataset was used to predict the values on test-set samples for drip loss%. The predicted drip loss% by the

prediction equation on the test-set samples resulted in 44% (n = 147) as superior and 11% (n = 37) as inferior. The predicted values and the observed drip loss% on the test-set samples are shown in figure 3. From the superior group 67 samples were predicted incorrectly as having more drip loss than 2% (e.g. 2.00% - 3.71%) while from the inferior group 23 samples were predicted incorrectly as having less than 4% drip loss (e.g. 2.71% - 3.96%). Therefore, from the superior group 118 out of 185 samples were predicted correctly (64%) and from the inferior group 29 out of 52 samples (56%) were predicted correctly.

Sorting on the 87 samples from the test-set of the production plant with the best prediction equation and most variation (CV 80%, plant A, Table 1) showed that from the superior group 72% was predicted correctly and from the inferior group 77% was predicted correctly. Table 5 shows the results from sorting with developed prediction equations for drip loss%. A lower variation at some plants might have negatively influenced the performance of the prediction equations. We found that only one inferior sample was predicted as superior (sample from plant D, true drip loss% 4.01% and predicted 1.95%).

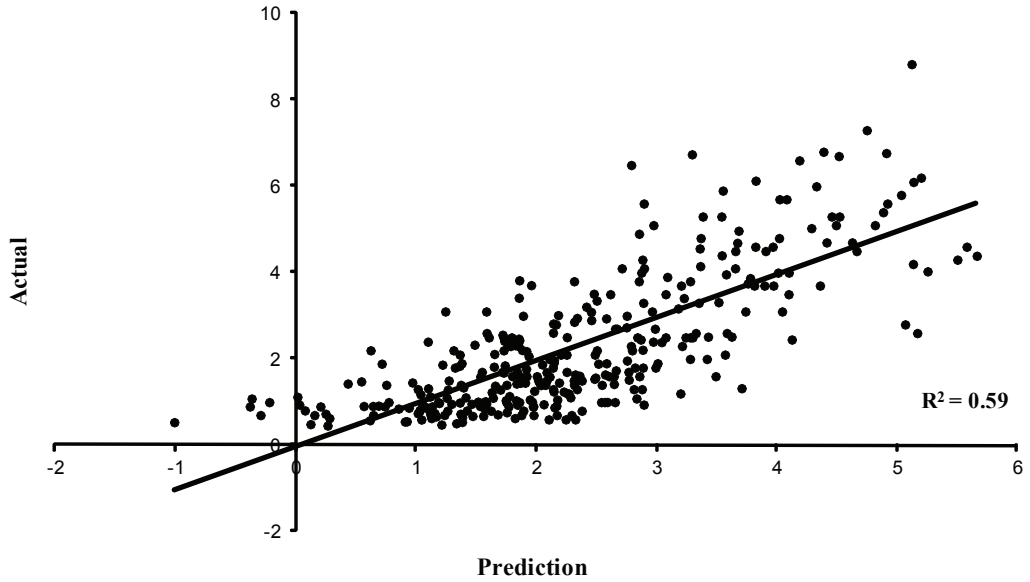

Figure 3. Predicted and actual measured drip loss% on test-set samples, using the prediction equation from the calibration-set derived from the combined dataset.

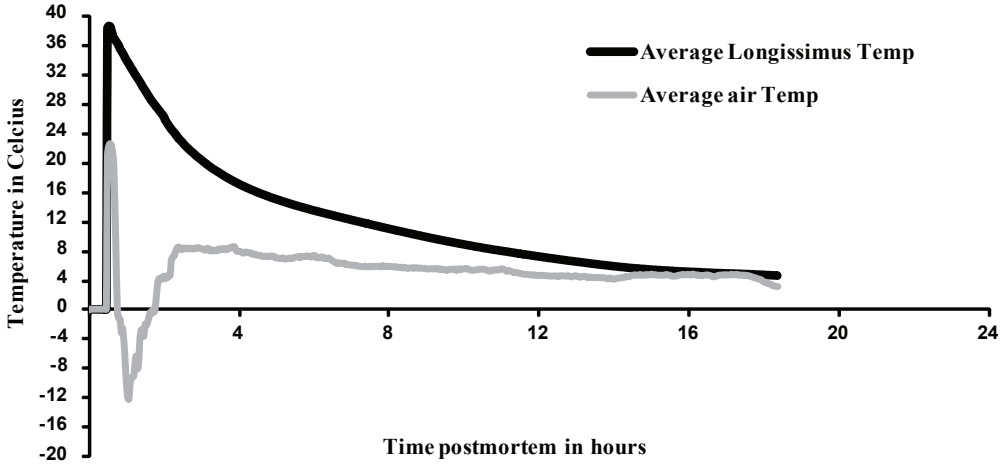

Figure 4. Average temperature curve of the *Longissimus dorsi* and air temperatures in plant A.

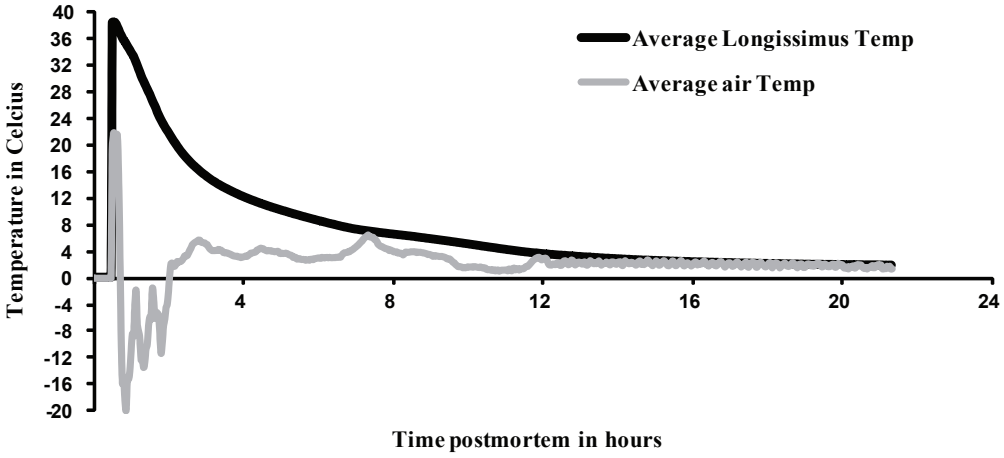

Figure 5. Average temperature curve of the *Longissimus dorsi* and air temperatures in plant B.

3.5 Prediction equations

The amount of ranking (factors), which was needed to develop prediction equations, varied between three and nine. $R^2 > 0.70$ and RPD > 3 was considered as a reliable prediction equation, while $R^2 > 0.70$ and 2 > RPD < 3 was seen as a useful prediction equation, which still needs improvement (Williams, 2007). For drip loss% the observed RPD values were ≥1.49 and ≤ 2.05, for colour L* the observed RPD values were ≥ 1.14 and ≤ 1.82 and pHu showed ≥ 1.12 and ≤ 1.57.

Data from production plant A showed relative high correlation coefficient (R^2 0.76) and RPD (2.05), while data from other plants showed $R^2 < 0.70$ and RPD < 2.0 for drip loss%. Plant A showed also highest variation (CV 80%) for drip loss%, which might have positively contributed to the development of a prediction equation in comparison to other plants. Equations on samples measured at pig processing plants could be developed with similar results as found in other studies (Barlocco et al., 2006; Brøndum et al., 2000; Garcia-Rey et al., 2005; Geesink et al., 2003; Hoving-Bolink et al., 2005; Leroy et al., 2004; Prieto et al., 2009; Rodbotten et al., 2000; Savenije et al., 2006) for drip loss% and colour L*.

Table 5. Sorting with the developed prediction equations was evaluated with the number of samples counted in the superior and inferior group. The number of samples was compared with the predicted values on these samples.

Location	Superior < 2.0 % drip loss% (n)	Correct %	Inferior > 4.0% drip loss% (n)	Correct %
A	54	72%	13	77%
B	64	89%	3	0%
C	33	48%	14	57%
D	30	57%	24	46%
Combined data	185	64%	52	56%

Highest correlation value for the prediction equation of colour L* was found with plant C. However, the results did not meet the requirements for screening at a production plant (R^2 0.70 and RPD 1.82). For the combined dataset, the prediction equation for drip loss% and colour L* showed a lower R^2 and RPD than the best prediction equation from plant A and C. The observed R^2 and RPD for pHu showed a coefficient of determination < 0.70 and RPD < 2.0, which indicates that the prediction equations cannot be used for useful screening on commercially slaughtered pigs. Drip loss% and colour L* were developed with a ranking lower than 10. A ranking below 10 indicates that over fitting did not occur. Hoving-Bolink (2005) stated that a large scanning area is necessary to reduce the effects of the variation of pork and improve the prediction equations (Hoving-Bolink et al., 2005). The Bruker device measured a area of approximately 4.5 cm^2 and covered the loin area of the sample. In this experiment we presented each sample once to the NIRS device. Every single sample presentation resulted in 32 scans. Presenting the sample more than once may have improved the results, while it also would have increased the total scanning time, which is a critical limitation for a practical application.

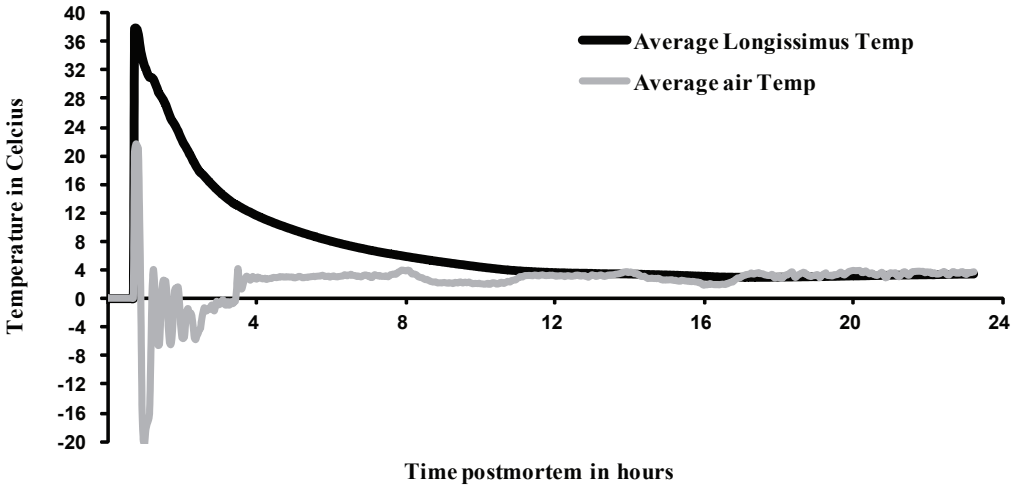

Figure 6. Average temperature curve of the *Longissimus dorsi* and air temperatures in plant C.

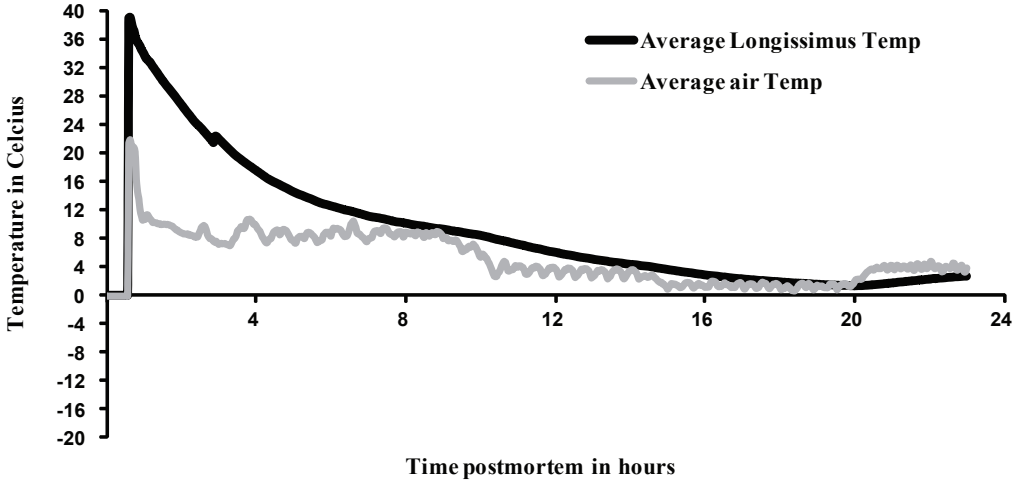

Figure 7. Average temperature curve of the *Longissimus dorsi* and air temperatures in plant D.

4. Conclusions

The observed R^2, RPD, RMSEE and RMSEP for drip loss% and colour L* indicated that prediction equations could be developed, on samples measured at production plants. The results for drip loss% and colour L* were comparable with results from other studies. Only the prediction equation for drip loss%, based on data from production plant A could be used for screening into three quality classes ($R^2 > 0.70$, RPD ≥ 2.0).

Prediction equation for colour L* developed from data of plant C showed highest R^2 and RPD values. However, the results from plant C did not meet the requirements for screening at a production plant (R^2 0.70 and RPD 1.82). pHu resulted in a $R^2 < 0.70$ and a RPD < 2.0 and did therefore not generate applicable prediction equations. The best prediction equation (plant A) for drip loss% was developed on data with the highest CV% (80%) and suggests that more variation might have improved the prediction equations for the other plants. The results of plant A are based on samples from commercially processed pork and can be considered as useful for screening and sorting for meat quality (drip loss%) at production locations. The production plants showed differences in cooling and carcass handling. The results suggest that prediction equations can be used for screening for drip loss% and should be developed per individual production plant.

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Table 4. Prediction equations obtained for the prediction of colour L*, pHu, and drip loss% in commercially processed pigs, by using an independent test-set validation.

				Calibration				Test-set validation			
Parameter											
Drip loss (%)	Location	Treatment	n	R^2	RMSEE	RPD	Rank	R^2	RMSEP	RPD	Bias
	A	A	174	76.3	0.89	2.05	7	76.1	0.81	2.05	-0.07
	B	A	182	65.8	0.80	1.71	8	53.9	0.73	1.49	0.11
	C	A	156	62.8	0.94	1.64	5	61.6	0.89	1.63	-0.11
	D	A	180	68.5	1.06	1.78	5	60.2	1.12	1.61	-0.20
	Comb	A	665	57.7	1.11	1.54	9	58.6	1.02	1.56	-0.05
Colour L*	A	C	174	67.2	3.37	1.75	6	55.4	3.6	1.5	-0.35
	B	A	182	48.1	3.52	1.39	5	22.8	3.53	1.14	0.32
	C	G + C	157	56.2	3.49	1.51	5	69.2	2.82	1.82	-0.44
	D	E	180	55.7	2.73	1.5	3	54.6	2.83	1.52	-0.64
	Comb	A	674	57.7	3.41	1.54	9	50.2	3.4	1.42	-0.12
pHu	A	E	174	38.7	0.15	1.28	6	39.5	0.13	1.29	-0.0
	B	F	182	65.0	0.1	1.69	9	26.7	0.13	1.17	0.01
	C	G	157	36.9	0.1	1.26	2	30.9	0.11	1.21	0.02
	D	G + C	152	49.3	0.09	1.4	7	34.8	0.10	1.24	0.01
	Comb	B	674	37.1	0.13	1.26	8	18.2	0.13	1.12	-0.02

Regions are expressed in reciprocal centimetres cm^{-1}. Treatment A is no spectral data pre-processing, treatment B is constant offset elimination, treatment C is straight line subtraction, treatment D is vector normalization, treatment E is min- max normalization, treatment F is multiplicative scatter correction, treatment G is 1st or 2^{nd} derivative with smoothing, treatment H is 1^{st} derivative with multiplicative scatter correction.

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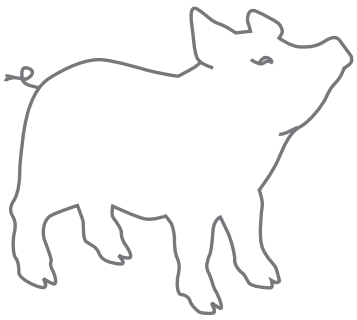

Chapter 3

Moisture absorption early post mortem predicts ultimate drip loss in fresh pork

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Abstract

Water holding capacity is the ability of meat to hold moisture and is subject to events occurring early post mortem in muscle tissues. The objective was to characterize the loss of moisture from muscle post mortem and investigate whether these losses are useful in predicting the ultimate drip loss% of fresh pork. Cotton-rayon absorptive-based devices were inserted in the *Longissimus* muscles of pork carcasses (n = 51) immediately post mortem and removed at 15 min intervals or remained in the muscle for 24 h. Drip loss% varied widely (0.6 – 15.3%) across carcasses. Greatest moisture absorption was observed at 105 min post exanguniation. Individual absorption at 75 min correlated with final drip loss% (r = 0.33). Correlations improved using individual absorption values at 90 min (r = 0.48) and accumulated absorption values at 150 min (r = 0.41). Our results reveal significant moisture is lost from muscle tissue early post mortem and suggest that select time periods post mortem, corresponding to the culmination of biochemical and physical events facilitating moisture release can be used for early drip prediction. Further, these studies suggest an approach for capturing moisture release early post mortem is possible and may be used to improve understanding or predict final drip loss% in fresh meat.

Keywords: Water holding capacity, drip loss%, pork

1. Introduction

Water holding capacity (WHC) is the result of biochemical physical changes occurring in muscle tissues post mortem and is largely influenced by live animal stress, genetics, pre-slaughter handling conditions and carcass cooling (Andres et al., 2007; Bowker et al., 1999; Rosenvold & Andersen, 2003). Water accounts for approximately 75% of the weight of meat (Offer, 1988a; Schäfer et al., 2002) and the ability of muscle to retain moisture is key to many meat quality parameters held in high regard by the industry and consumers (Huff-Lonergan & Lonergan, 2005; Schäfer et al., 2002). Much effort has been invested in understanding the mechanisms of how moisture is entrapped in the tissues and is subsequently lost during its conversion to meat. Moisture is retained in muscle as intra- and extra cellular moisture and is bound to varying extents in these locales (Huff-Lonergan & Lonergan, 2005). Larger amounts of unbound, or less tightly associated water, are often associated with reduced water holding capacities in fresh meat, and as such, it is widely accepted that this type of moisture is responsible for drip loss%.

The aforementioned, which change during periods of significant energy metabolism and rigor development, influence WHC. Traditionally, WHC is either measured as drip loss% or ultimately observed as purge in fresh meat packaging. Efforts to predict WHC include a number of techniques that include applying various forces on meat to liberate moisture from the tissues (Christensen, 2003; Reiner Hamm, 1961; Honikel, 1998; Kauffman, 1993). Regardless of the method, most all analyses begin traditionally at or after a 24 hr post mortem chill, which limits the utility of such information to sort carcasses. Therefore, the objective of our work was to characterize the loss of moisture from muscle post mortem and investigate whether these losses are useful in predicting the ultimate drip loss% of fresh pork.

2. Materials and Methods

A dataset from 51 pigs, harvested over six days at two different locations (location A and location B) were used in this study. Crossbred pigs (n = 10) weighing approximately 100 kg were transported to the Purdue University Meat Science Research and Education Center (location A). Pigs were slaughtered over a two day period using standard industry practices. After electrical stunning, carcasses were scalded, split and held at room temperature (~20 ˚C) for 60 min before being placed in the chill cooler (4 ˚C) overnight. The facility operated under regulations established by the United States Department of Agriculture, Food Safety and Inspection Service, and was in compliance with the Humane Slaughter Act. The remaining pigs (n = 41) were slaughtered at the Virginia Tech Meat Center (location B) during three slaughter days. These pigs consisted of commercial crossbred pigs harboring the Halothane and Rendement Napole genes. Pigs were processed using similar protocols as used at location A, with the exception that the pigs were stunned twice. Moreover, to increase the variation of drip loss% within this population of pigs, five carcasses were randomly subjected to an electrical stimulation protocol at location A as outlined by Bowker et al., (1999).

2.1 Muscle pH values

At location A, pH values were recorded in the *Longissimus* muscle (LM) between the 10th and 11th rib at 45 and 180 min and 24 h post mortem using a Beckman Φ110 ISFET pH measuring device (Beckman Instruments, Inc., Fullerton, CA USA) equipped with a spear-tipped KCL^{-} Φ Smart ISFET standard probe (Beckman Instruments, Inc., Brea, CA USA). At location B, muscle samples were also collected from the LM at 10th and 11th rib at 45 and 180 min and 24 h post mortem and immediately frozen in liquid nitrogen. Frozen samples were ground and approximately 1.0 g was placed in an iodoacetate solution (5 mM sodium iodoacetate, 150 mM potassium chloride; pH 7.0) and pH was measured.

2.2 Moisture Absorption

At 15 min post-stunning, a 14 mm diameter coring device was used to create a hole in the LM approximately 6.5 cm deep at the juncture of the 3rd and 4th lumbar vertebrae. Approximately two g of cotton-rayon material (location A: O.B. regular absorbency; Johnson & Johnson, Montreal, Quebec Canada; 4.5 cm long x 1.2 cm diameter; location B: Tampax, Tambrands Manufacturing Inc., Auburn USA; 4.5 cm long x 1.2 cm diameter) was fully inserted into the hole. All sampling was performed in duplicate and averaged. Absorptive materials were weighed before and after the sampling. Sampling began at approximately 15 min after exsanguination. Absorptive materials were replaced with new absorptive materials after a 15 min dwell time. Sampling continued until 300 min post mortem, at which time new absorptive material was placed in the core and allowed to remain in the LM until 24 hr post mortem. Time post mortem was used to designate time at which material was removed and weighed. Individual absorption values were calculated for each time point. Accumulated absorption values equal the sum of absorption values at each consecutive time point. Moisture absorbed was also normalized against the total moisture absorbed.

2.3 Prediction of drip loss by absorption at 24 h post mortem

Twelve carcasses were used to determine whether drip loss% could be predicted at 24 hr by the aforementioned moisture absorption technique. Absorptive material was inserted in the *longissiumus dorsi* at nine different locations along the loin at 24 hr post mortem. Sampling locations began at the *longissiumus* muscle opposite the 10th rib and continued every 2.54 cm to a location opposite the last lumbar vertebra. Dwell times were 2, 5, 15, 30, 60 or 90 min. Sampling for each dwell time was conducted in triplicate and averaged for a single value. Absorption was calculated as average weight gain per dwell time.

2.4 Drip loss

Drip loss% was determined using the Danish drip tube technique (Rasmussen & Andersson, 1996). Two samples for drip loss% were taken at 24 hr post mortem from every carcass used for these experiments. A 2.5 cm thick slice chop was removed adjacent to the 10^{th}-11^{th} ribs. Triplicate core samples were removed from the *Longissmus* muscle of chops using a 2.5 cm-diameter coring device. Cores were placed in drip loss tubes and then allowed to equilibrate for 24 hr at 3 ˚C. All drip loss% values for each carcass were averaged. Drip loss% was determined as weight loss percentage of initial sample weight. All data of both locations were combined to generate a combined dataset with maximal variation on the parameters.

2.5 Statistical analysis

All data were subjected to the PROC procedures of SAS (SAS® 9.1 Inst., Inc., Cary, NC). The carcass was used as the experimental unit. PROC GLM, PROC REG and PROC CORR procedures of SAS were used to determine relationships between absorptions and drip loss% values. During analysis, carcasses were grouped on their drip loss value (0 – 5%, 5 – 10%, 10+%) to allow comparisons between the level of drip loss% of carcasses and the levels of absorptions. The groups were arbitrary chosen.

3. Results

3.1 Moisture and early post mortem changes in muscle

The calculated Pearson correlation coefficients of the absorption values (individual, accumulated, and normalized) are expressed in table 1. Individual weight absorbed at 15 min dwell time correlated ($P < 0.05$*)* with drip loss% from 75 until 120 min, 240 until 270 min and 300 min until 24 hr post mortem. Accumulated absorption correlated ($P < 0.05$*)* with drip loss% from 90 min until 300 min post mortem and from 300 min until 24 hr dwell time ($P < 0.05$). The greatest correlation was observed at 90 min post mortem ($r = 0.48$, $P < 0.01$) for individual absorption and 300 - 1440 minutes for the accumulated absorption ($r = 0.50$, $P < 0.01$). No significant correlations were found between individual or accumulated absorption values and drip loss% during the first 75 min. To understand better how moisture may be released over time from muscle tissues post mortem, absorption was normalized and expressed as a percentage of total moisture absorption over the first 24 hr after slaughter. Results revealed that normalized absorption values from 30 - 90 min, and from 165 - 1440 min correlated ($P < 0.05$) with drip loss%. Absolute absorption values were greater ($P < 0.05$) for carcasses with high drip loss% values than for carcasses with lower drip loss% values.

Figure 1 shows the mean absolute 15-min absorption values or moisture released from the

muscle tissue with time post mortem. Moisture absorption increases from muscle between 45 to 105 min post mortem. A decline in absorption was observed after the peak at 105 minutes. The maximum amount of water absorbed from muscle peaked between 60 and 105 min post mortem for all carcasses in the study (data not shown).

Normalized moisture release data are summarized by drip loss% category (Table 2). A significantly greater proportion of moisture was lost ($P < 0.05$) from the low drip loss% pork between 30 and 90 min, and 195 and 300 min post mortem compared to the high drip loss% category, while the medium drip loss% group was intermediate. By 105 min post mortem, statistically significant differences between the drip loss% groups disappeared but returned ($P < 0.05$) after roughly one and one half hours, at 195 min post mortem (Table 2). At 195 min, similar differences were noted ($P < 0.05$) among drip loss% categories as observed for moisture collected between 30 and 90 min post mortem. In addition, over half the moisture absorbed from muscle tissues was available by 300 min post mortem across all drip loss% categories. A greater release ($P < 0.05$) of moisture was observed in high drip loss% group in comparison to the low drip loss% group during the remainder of the post mortem period (300 – 1440 min), table 2. Thus, the amount of total accumulated absorption was different ($P < 0.001$) between the high and low drip loss% groups (0 – 5% drip loss: 4.7 grams, 5 – 10% drip loss: 9.3 grams, 10 – 15% drip loss: 9.8 grams), and the medium drip loss% group was different from the low ($P = 0.0007$) but not different from the high ($P = 0.62$) group.

3.2 Absorption at 24 hours post mortem

Dwell time affected ($P < 0.001$) moisture absorption (Table 3). Mean drip loss% for all samples was 5.90%. Absorptive material inserted for only two min absorbed the least amount of moisture (0.07 g) compared to that inserted for longer periods of time (30, 60, and 90 min). Absorption values for 30 min dwell time (1.51 g) was smaller ($P < 0.05$) than values for from 60-minutes dwell time (1.93 g) and a 90-minutes dwell time (2.24 g). A correlation between absorption for a 15-min dwell time ($r = 0.84$, $P < 0.001$) and drip loss% was greater than correlations of the other dwell times to drip loss% (0.62 to 0.81).

3.3 pH

Measured pH values of the combined dataset at each time post mortem, were grouped by drip loss% and are summarized in Table 4. Though not significant at all time points, lower pH values coincided with a higher drip loss% category. Pearson correlation coefficients between pH and drip loss% revealed significant correlations between pH at 45 min ($r = -0.34$, $P = 0.01$), 180 min ($r = -0.48$, $P < 0.001$) and 24 h ($r = -0.47$, $P < 0.001$). Analysis revealed that pH measured at 24 h post mortem explained 23% of the variance in drip loss%, and pH measured at 180 min post mortem explained 23% of variation. In contrast, pH measured at 45 min post mortem only explained 11% of the variance in drip loss%.

Table 1. Pearson correlation coefficients for drip loss% and individual absorption, accumulated and normalized (% total) absorption values over time post mortem.

	Absorption		
Time (minutes)	Individual[1] (g)	Accumulated[2] (g)	Normalized[3] (%)
30	0.13	0.13	-0.53**
45	-0.08	-0.03	-0.59**
60	0.19	0.08	-0.54**
75	0.33*	0.2	-0.50**
90	0.48**	0.35*	-0.37**
105	0.34*	0.40**	-0.27
120	0.33*	0.39**	-0.25
135	0.27	0.39**	-0.25
150	0.25	0.41**	-0.24
165	0.06	0.39**	-0.28*
180	0.07	0.37**	-0.31*
195	0.22	0.37**	-0.33*
210	0.09	0.36**	-0.36**
225	0.25	0.36**	-0.40**
240	0.37**	0.37**	-0.42**
255	0.39**	0.37**	-0.43**
270	0.33*	0.38**	-0.45**
285	0.19	0.37**	-0.48**
300	0.22	0.38**	-0.50**
300- 1440[†]	0.58**	0.50**	0.50**

**P < 0.05 **P < 0.01*

[1] Correlation coefficients between drip loss% and 15 minutes absorption values measured at various 15 minutes intervals post mortem.

[2] Pearson correlation values between drip loss% and the accumulated absorption values from cotton material inserted for 15 minutes in the meat.

[3] Pearson correlation values between drip loss% and the accumulated absorption values at each time point, which are expressed as percentage of total absorption during 24 hours.

[†]Absorptive material inserted at 300 minutes and left until 24 hr post mortem.

Table 2. Percentage total moisture absorbed until 24 hrs post mortem, compared with absorption percentage per minutes post mortem, sampled in *Longissimus dorsi* of carcasses with 0 – 5 %, 5 – 10%, and 10+ % drip loss %.

	Drip loss category (%)		
Time (minutes)	Low 0 – 5% (n = 11)	Medium 5 – 10% (n = 18)	High 10+% (n = 22)
30	7.3	4.3[a]	3.3[a]
45	12.5	7.5[a]	5.4[a]
60	17.5[a]	12.5[ab]	9.2[b]
75	22.2[a]	18.7[ab]	13.7[b]
90	26.8[a]	24.2[ab]	19.6[b]
105	31.8	30.2	26.4
120	36.9	35.4	31.7
135	41.3	39.8	35.9
150	45.4	43.0	39.9
165	49.1	47.1	43.0
180	52.5	50.0	45.3
195	55.7[a]	52.6[ab]	47.9[b]
210	58.6[a]	55.0[ab]	49.8[b]
225	61.1[a]	57.0[ab]	51.5[b]
240	63.4[a]	59.1[ab]	53.5[b]
255	65.5[a]	60.8[ab]	55.4[b]
270	67.7[a]	62.3[ab]	57.1[b]
285	70.1[a]	64.0[ab]	58.6[b]
300	72.2[a]	65.7[ab]	60.0[b]
300 - 1440[†]	27.8[a]	34.3[ab]	40.0[b]
LSMeans	44.28[a]	41.19[a]	37.36[b]
SE	1.36	1.06	0.96

[†]Absorptive material inserted at 300 minutes and left until 24 hr post mortem.
The values in the table are expressed as percentage (%) of total absorption during 24 hours. [ab] Means within the same row, bearing different letters are different (P < 0.05).

4. Discussion

4.1 Moisture absorption and early post mortem changes in muscle

Moisture is readily absorbed from meat when it is loosely bound (Hamm et al., 1961). Essentially, water is held in muscle tissue by chemical bonding, hydrogen bonding or by capillary forces. Chemically bound water or water that is bound strongly to charged proteins is not easily released from muscle and represents a small proportion of total water in muscle (Huff-Lonergan & Lonergan, 2005). Water held by negatively charged proteins becomes less bound when positively charged ions, like hydrogen, increase during post mortem metabolism due to ATP hydrolysis (Bendall, 1962). During the early post mortem process, significant reductions in ATP level, aggregation and denaturation of muscle proteins most likely cause pre-rigor sarcomere shortening and the formation of drip channels in the muscle that enlarge interstitial spaces for water to escape the structure (Honikel et al., 1986; Lawson, 2004). As a result, less tightly bound water accumulates in muscle and migrates to interstitial spaces (Offer, 1991).

Data in table 1 reveal that the amount of moisture absorbed continued to increase between 30 and 105 minutes post mortem, after which a decline in water loss was observed. A continuous increase in the absorption of moisture until 105 min argues the aforementioned processes whereby accumulated hydrogen ions are expected to reduce binding of hydrogen bound water indeed occurs (Bendall, 1962). The rate of the pH decline post mortem appeared to parallel the water binding in meat and corresponded with observed positive correlation values ($r = 0.07 – 0.58$) between absorption and drip loss% in table 1 (Fischer, 2007; Honikel et al., 1986). The pH values in table 4 indicate that indeed carcasses with high drip loss% had a more rapid pH decline at 45 min (pH 6.05) and 180 min (5.44) post mortem than carcasses with low drip loss% (pH 6.21 at 45 min, pH 5.65 at 180 min). Although, pH values were significantly different between low (pH 5.65) and high (pH 5.44) drip loss% grouped carcasses at 180 min, pH values at 45 min were not significant different, table 4.

One post mortem event known to influence water binding in meat occurs as early as 45 min and involves proteolytic degradation of desmin, an intermediate filament that acts to tether myofibrils in register (Huff-Lonergan & Lonergan, 2005; Melody et al., 2004). Any changes in desmin, or closely associated proteins could alter the lateral spacing of myofibrils within a muscle cell and could dramatically impact moisture held in this location. However, no significant correlations were found between moisture absorption and drip loss% at 45 min in table 1. At 45 min, significant amounts of ATP are still available in the muscle tissue and as such, continued hydrolysis could result in greater hydrogen production and ultimately result in a significant loss of bound water, similar to that found in table 1 between 75 and 90 min (Batlle et al., 2001; Bendall, 1951). Even so, the bulk of hydrogen produced before 75 min should still be largely buffered by either the inherent buffering capacity of the tissues or the inorganic phosphate being generated by additional ATP hydrolysis (Puolanne & Halonen, 2010). Hydrogen ion accumulation indirectly results in the formation of water channels (Honikel et al., 1986; Lawson, 2004). Further, muscle proteins collapse as the pH in

the environment drops closer to the isoelectric point of muscle, which is where the net charge among the various muscle proteins is close to zero (Cassens et al., 1975; Honikel, 1998; Offer, 1991).

Figure 1 reveals that the maximum levels of absorption were found at 105 min, which might be related to the aforementioned collapse of muscle proteins. However, the maximum absorption was expected to occur at later times (> 3 hours post mortem) (Offer, 1991). If moisture loss from muscle early post mortem was due solely to pH-related phenomena, moisture should continue to exude from the muscle until the ultimate pH of the meat is reached, and ultimate pH is generally considered to occur at 24 hrs post mortem, table 4 (Eikelenboom et al., 1974a). There is little question that the rate and extent of post mortem muscle metabolism especially influences water binding early post mortem (Enfalt et al., 1993; Huff-Lonergan & Lonergan, 2005). However, if this is true, continuous moisture should emanate from the tissues until rigor is completed (Kristensen & Purslow, 2001; Schäfer et al., 2002). Data from table 1 and figure 1 indicate that the moisture release indeed continued to at least five hours post mortem. However, given that maximum absorption occurred in the vicinity of 105 min post mortem (Figure 1), rigor would be expected to occur very soon on either side of the event. Given the most have documented rigor at approximately three hours post mortem (180 minutes) this seems unlikely (Dransfield & Lockyer, 1985; Offer, 1991), however, Honikel et al., (1986) suggested that rigor onset begins at a pH of 6.1 to 6.2 rather than a specific time post mortem. According to our pH values (Table 4), carcasses used in these experiments may have experienced the onset of rigor at 45 min post mortem which agrees with the observed increase of absorption up to 105 min post mortem (Honikel et al., 1986).

At location A, five carcasses were subjected to a non-natural electrical stimulation event, and thus were expected to generate an increased post mortem metabolism. Electrically stimulated carcasses had significant ($P < 0.05$) higher drip loss% values (+3.7% drip loss) than the carcasses which were not subjected to electrical stimulation, and agrees with results from other studies (Anil & McKinstry, 1998; Bowker et al., 1999; Hammelman et al., 2003).

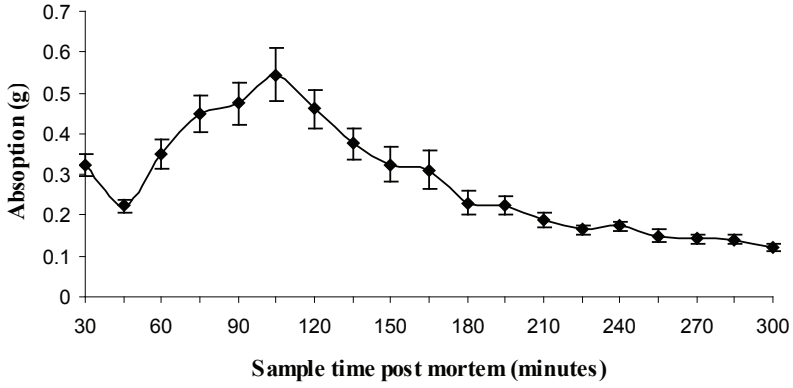

Figure 1. Average moisture absorption (g) at different times (minutes) post mortem. Sample time post mortem (Petersen et al., 2005) reflects time at which absorptive material was removed from the muscle and weighed. Date presented as means ± SEM within time.

It is known that rigor onset coincides with ATP depletion and once this process is complete, changes in protein structure should cease and stabilization of the muscle structure would result in slower water liberation (105 – 225 min post mortem, Figure 1). Indeed at later stages (>200 min post mortem), we observed that absorption of moisture by cotton-rayon material was reduced to approximately 0.15 g or less for all samples (Figure 1). This trend continued through 300 min (5 hr) post mortem arguing rigor causes liberation of moisture loss in muscle early post mortem (105 min), a theory not completely held by all (Honikel et al., 1981; Kim et al., 1985). Regardless, data showed a greater proportion of moisture loss from muscle tissue early post mortem (within 2 hrs), followed by a period of reduced output (105 – 225 min), followed by period of relatively stable release of moisture (240 – 300 min). We expect that this rather stable phase of water release would continue until effects of post mortem proteolysis are realized, at which time this would increase WHC of the meat by breakdown of cytoskeleton structures and creating intramyocellular spaces for water loss (Kristensen & Purslow, 2001).

Accumulated absorption values revealed that from 45 min post mortem onward greater moisture is liberated from muscle tissues ultimately creating meat with high drip loss%. Differences between accumulated absorption become more pronounced with time post mortem and may account for differences in absorption at the end of the first 24 hr post mortem. The normalized values in Table 2 indicate that the relative absorption was significantly ($P < 0.05$) greater (40% versus 28%) for carcasses with high drip loss% (10+%) than for carcasses with low drip loss% (< 5%) after 300 min post mortem.

Table 3. Average moisture absorption (g) at 24 hrs post mortem for each dwell time (minutes; n = 12) and drip loss (%; n = 12).

Dwell time (minutes)	Mean (g)	SD	Minimum (g)	Maximum (g)	CV (%)	r
2	0.70 [e]	0.23	0.39	1.17	32.5	0.62*
5	0.85 [de]	0.28	0.56	1.34	33.0	0.81**
15	1.07 [d]	0.37	0.56	1.70	34.5	0.84**
30	1.51 [c]	0.50	0.75	2.27	33.2	0.79**
60	1.93 [b]	0.64	1.04	2.98	32.9	0.81**
90	2.24 [a]	0.72	1.07	3.54	32.3	0.79**
Drip Loss (%)	5.90	2.51	2.99	9.57	42.5	-

[ab] Means within the same column, bearing different letters are different ($P < 0.05$).

** $P < 0.05$; **$P < 0.01$; ***$P < 0.0001$.*

One potential pitfall associated with our experimentation is the unknown absorbency of our material. We assume for the sake of explaining our data that the water absorbed by the cotton material was available (unbound) water, present in the interface between the incision and the muscle. Clearly, a cellulose material like the cotton-rayon material is anionic, has water absorbing characteristics and has a capillary suction pressure (El-Naggar et al., 2006; Rowland & Howley, 1988). These hydrophobic properties of cotton-rayon material were expected to compete with the muscle's water binding ability. The cotton-rayon materials used in this study has a capillary suction pressure of approximately 80 mm Hg (Foley et al., 2004) and the capillary pressure of pork meat is estimated to 22.5 $x10^2$ mm Hg which is considerably larger than the suction pressure of the cotton-rayon material (Puolanne & Halonen, 2010; Trout, 1988). Therefore, the competing suction pressure of cotton-rayon was expected to be lower than the natural water holding of myofibrils and capillary spaces. The total absorption capacity of cotton-rayon material was approximately 6.8 g, and was therefore expected to be sufficient (highest observed individual absorption value was 3.0 g) for this experiment (Chatterjee & Spotswood, 1973).

4.2 Absorption measured at 24 hours post mortem

Results on the absorption dwell times, measured on meat at 24 hrs post mortem (Table 4) indicate that 15 min dwell time had highest correlation to drip loss% ($r = 0.84$, $P < 0.01$). Therefore 15 min dwell time was chosen throughout this experiment for the absorption measurements. It is not clear why dwell times longer than 15 min had lower correlations to drip loss%. One can assume that after the cotton-rayon material was inserted, the water migration requires time to equilibrate between the material and the meat. Dwell times shorter than 15 min might therefore have reduced correlations to drip loss%. On the opposite, dwell times longer than 15 min should have enhanced water migration from further locales in the meat, which can be affected by the physical properties, such as capillary pore size of the meat (Dullien et al., 1977). The capillary systems may have been different between carcasses, which theoretically could have reduced the correlations between drip loss% and absorption values from dwell times longer than 15 min. Regardless, the lack of increased correlations with dwell time remains unclear.

4.3 pH

In general, carcasses with higher drip loss% values often reveal an accelerated post mortem pH decline and a lower ultimate pH (pHu) than carcasses with lower drip loss% values (Bowker et al., 1999; Eikelenboom et al., 1974b; Hallund & Bendall, 1965; Hammelman et al., 2003; Schäfer et al., 2002). Results in table 4 showed that carcasses with low (0% - 5%) drip loss% indeed had higher pH than carcasses with high (10+%) drip loss% ($P < 0.05$), at both 180 min and 24 hours post mortem. It has been established for some time that considerable variation exits in rate and extent of post mortem muscle pH decline (Briskey, 1964). Major gene effects have been explicitly implicated

in a rapid (Halothane gene; Mickelson et al., 1988) and an extended muscle pH decline (low ultimate pH) (Rendement Napole gene; Milan et al., 2000). An additive effect of these genes is sometimes observed in pigs carrying both genetic mutations, which were used in this study and expected to create variation (Copenhafer et al., 2006). The rate of early post mortem pH decline affects the water binding capacity of the cytoskeletal proteins when the carcasses are still warm. Whereas the ultimate pH indicates the extent of the post mortem pH, which is not dependent of the rate of early post mortem pH decline. The results of this study underline that the rate of early post mortem pH decline and ultimate pH can be seen as two different mechanisms which affect WHC of the meat. The rate of post mortem pH decline might therefore explain the significant correlations found in this study between absorption at 75 minutes through 120 minutes post mortem and drip loss% values. The ultimate pH level might explain the significant correlations between absorption at 240 min through 24 hours post mortem and drip loss% in table 1.

Table 4. Mean pH values (±SEM) measured at 45 minutes, 180 minutes, and 24 hours post mortem from the Longissimus muscle of carcasses segregated by 24 hr drip loss category (0 - 5, 5 -10, 10+ %).

	Drip loss category					
Time post mortem	0 – 5%		5 – 10%		10+%	
	pH	SEM	pH	SEM	pH	SEM
45 minutes	6.21	0.10	6.17	0.07	6.05	0.04
180 minutes	5.65[a]	0.07	5.56[ab]	0.04	5.44[b]	0.03
24 hours	5.55[a]	0.06	5.53[a]	0.04	5.39[b]	0.02

[ab] *Means within the same row, bearing different letters are different ($P < 0.05$), SEM values indicate the standard error of the mean.*

5. Conclusions

Results of these studies demonstrate that maximal moisture released from pig muscle post mortem peaks within the first two hours after stunning. Over fifty percent of the total moisture was released within the first five hours post mortem in a drip loss-dependent manner, most likely in response to variations in metabolism and pH decline. Moreover, insertion of an absorptive material early post mortem into the muscle tissue of pork carcasses absorbs moisture and correlates to ultimate drip loss% values of fresh pork. These data suggest that early post mortem absorption can predict pork drip loss% of fresh pork at 24 hours post mortem, indicative of the overall WHC. Creation of an absorptive-based technology may be possible early post mortem that predicts WHC, a critical component of fresh meat quality.

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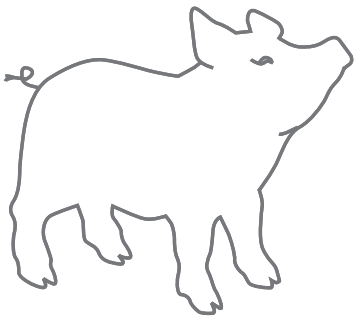

Chapter 4

Can near infrared spectroscopy (NIRS) be used to optimize technological yields in bacon production?

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Abstract

The aim of this paper was to investigate if near infrared spectroscopy (NIRS) could be used to optimize back bacon production and if NIRS prediction equations could be developed for prediction of technological yields of this type of bacon. Two experiments were conducted on middles from 356 and 329 carcasses and meat quality and processing yields were measured. Fresh meat quality measurements (pHu and colour L*) had low, but significant correlations ($r \leq 0.26$) with technological back bacon yields. Developed NIRS prediction equations for drip loss% resulted in low R^2 (0.50) while technological yields ($R^2 < 0.21$ and RPD < 1.1) could not be predicted. The pre-selected groups by NIRS yielded no significant differences on the fresh meat quality parameters and on injection yield and yield before frozen storage. Differences between both groups were not significant and were not regarded as relevant.

Keywords: back bacon, NIRS, WHC, fresh meat quality, technological quality

1. Introduction

The back bacon production consists of processing steps where a salt solution (brine) is injected to the meat to preserve it and create the specific sensory quality of the bacon (Eddy & Kitchell, 1961; E. Puolanne & Halonen, 2010). Technological yield values of back bacon are directly influenced during bacon production by adding brine to the meat, but also by the meats ability to retain the added brine and its own naturally occurring water content. The pork water holding capacity (WHC) has been reported to relate to the drip loss% of cured products like bacon (Fisher et al., 2000; Wismer-Pedersen, 1960). Sorting for pork primals based on their WHC at the start of the back bacon production might therefore be useful to optimize the technological yields during bacon production. Nowadays, the pork processing industry has been specialized in sorting on weight and lean characteristics of carcasses and primal cuts. Rapid sorting for WHC has not been achieved mainly due to the lack of rapid on-line non-invasive pork quality measurements devices.

Near infrared spectroscopy (NIRS) was identified as a potential technique that could allow sorting of pork primals according to different WHC categories (Barlocco et al., 2006; Geesink et al., 2003; Kapper et al., 2012; Prieto et al., 2009; Swatland & Irie, 1992). NIRS is based on the principle that components (O-H, C-H, C-O, N-H) in organic molecules absorb or emit NIR frequencies when their vibrational state changes, which can be used to identify or predict properties of the products (Cen & He, 2007). Kapper et al. (2012) concluded that NIRS could be used to select on predicted pork WHC under commercial cold room conditions (Kapper et al., 2012). The objective was to investigate if NIRS prediction equations are relevant for prediction of technological yields during back bacon production.

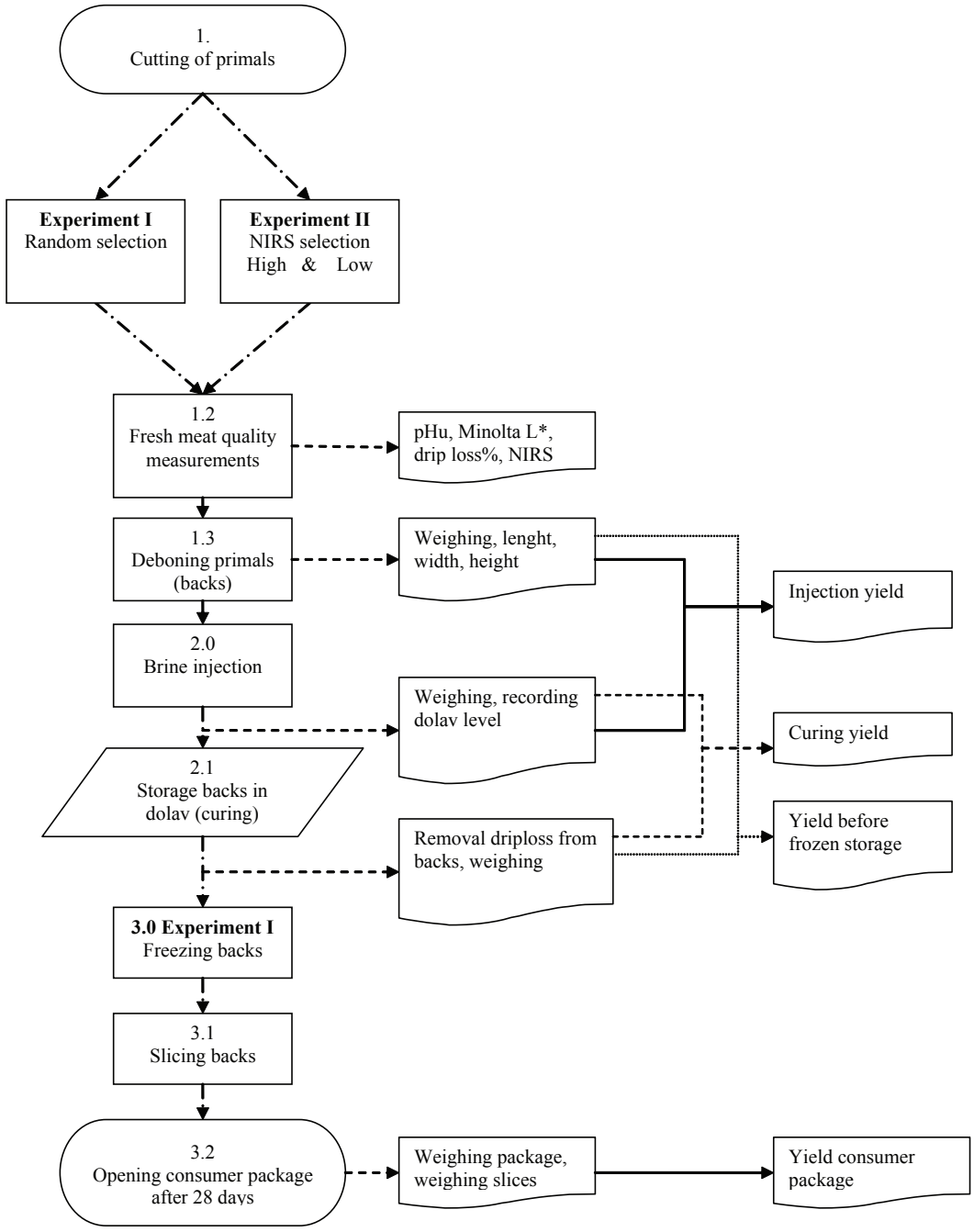

Figure 1. Flow chart of experiment 1 and experiment 2, including description of measurements during the back bacon production.

2. Materials and methods

2.1 Experimental set-up

Two experiments were carried out at two Dutch pork processing plants. Both plants used electrical stunning of pigs prior to exsanguination and bleeding. Middles for back bacon production were selected from carcasses with an average weight of 92 kg, in the cutting room at 24 hours post mortem. The process flow chart of the bacon production and measurements is displayed in figure 1.

2.1.1 Experiment 1

A total of 356 middles (*Longissimus dorsi*) were collected from randomly chosen carcasses during one day. A NIRS measurement on the fresh cut surface of a slice from the *Longissimus dorsi* was taken in the cold room (~4 ˚C). The contact free NIRS probe was placed on a tripod and measurements were made in downward direction (Figure 2). NIRS spectra were measured with a Bruker Matrix-FE device (Bruker Optics GmbH, Ettlingen, Germany) using a contact free probe (Q-412 Bruker Optics GmbH, Ettlingen, Germany) with an integrated light source. The device measured reflectance from 4.000 to 12.000 cm^{-1}. The NIRS spectrum was automatically transformed into Log 1/R absorption spectrum.

A total of 32 NIRS scans were averaged per sample with a chosen wavelength resolution of 32 cm^{-1}. The NIRS scanning time was approximately 3.8 seconds per sample. Specific NIRS prediction equations for bacon technological quality were made.

2.1.2 Experiment 2

A total of 329 middles (*Longissimus dorsi*) were collected from pig carcasses during two consecutive days. The middles were divided into two different drip loss% groups (Low and High) by predictive NIRS measurements according to prior developed prediction equations for WHC (Kapper, Klont, Williams et al., 2012), since the results from experiment 1 were inconclusive to develop bacon specific technological yield prediction equations. Middles with a predicted drip loss% value < 1.47 % were classified as Low and those with predicted drip loss% value > 3.14 % as High. NIRS scans were taken directly on the fresh cut surface of the middle from the shoulder side. The NIRS scanning time was approximately five seconds and was set to 16 cm^{-1} and 16 scans were taken at a scanner velocity of 10 KHz. Scanning settings were almost similar to the prediction model conditions of Kapper et al. (2012), except that in Kapper et al. (2012) 32 scans were taken.

Figure 2. Measurement setup of the contact free probe and NIRS device.

2.2 Meat quality measurements

The meat quality measurements were collected in the cutting room of the plant and the sampling was done at approximately 24 hours after exsanguinations. Slices of approximately two cm thick were taken from the *Longissimus dorsi* at the shoulder side from the middle at the height of the 4th rib. Colour L*, a*, and b* colour values (Chroma Meter CR-400, Konica Minolta Sensing Inc., pulsed xenon lamp, diffuse illumination and silicon photocells detector) were measured after a minimum of 30 minutes blooming. Ultimate pH (pHu) was measured in the bone-in loin (10th rib) with a MPI pH-Meter (Meat Probes, Inc). The pork slices were weighed and individually stored at approximately 3°C during 48 hours in consumer retail trays to measure drip loss% over 48 hours (Kapper et al., 2012). Sampling procedures are described in Kapper et al. (2012).

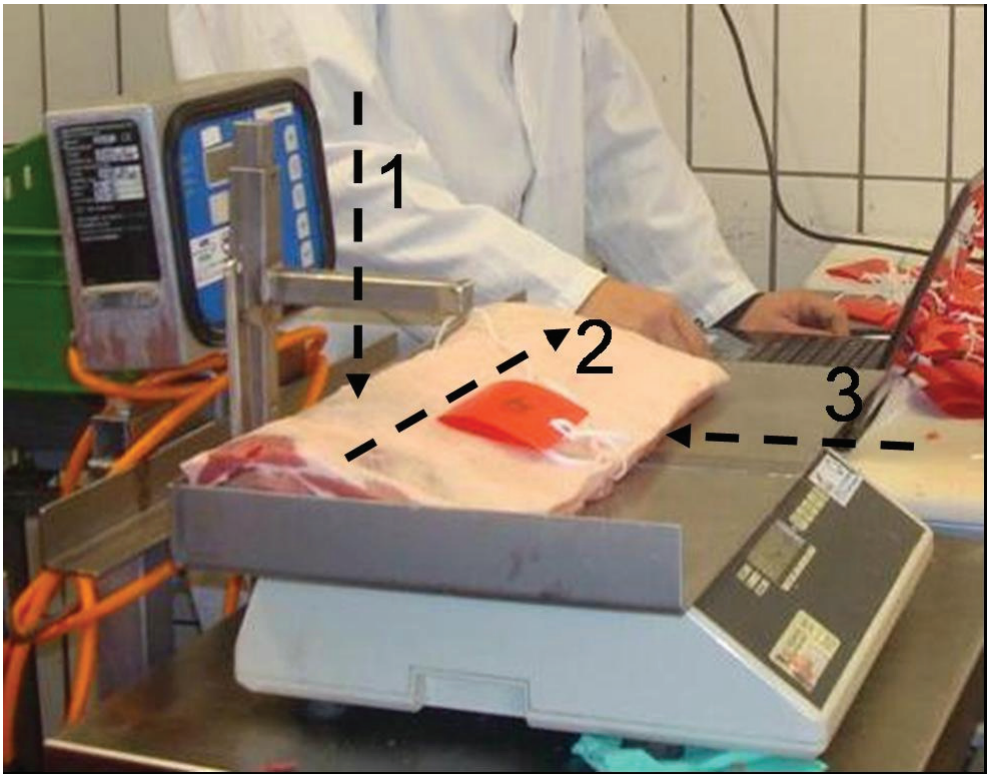

Figure 3. Measurement of the labelled and de-boned backs, height (1), length (2), and width (3).

2.3 Yield measurements during bacon production

All selected middles were individually labelled and stored at 3°C until next day's transport to a bacon production plant within The Netherlands. At approximately 72 hours post mortem, the middles were de-boned and cut according to the specifications of commercially produced back bacon for the UK market. The prepared samples (backs) were labelled and weighed directly after de-boning. Length (cm), width (cm), and height (cm) were recorded for every sample with a standardized measuring device (Figure 3) and were transferred to the brine injection device. The back bacon was produced with 3.4% salt. Brine components were; water, NaCl, preservatives (Sodium Nitrite E250 ($NaNO_2$), Potassium Nitrate E252 (KNO_3)), anti oxidants (Sodium Ascorbate, E301, ($C_6H_7O_6Na$)). Samples were randomly transferred into the injection equipment. Following injection, the backs were immediately tumbled in-line for approximately two minutes. All backs were individually weighed after tumbling and the final injection yield was calculated.

Injection yield was calculated as injection yield $= \frac{W2}{W1} x100\%$

W1 = weight of the back bacon before injection

W2 = weight of the back bacon after injection and tumbling

After injection, all backs were vacuum packed in plastic bags and stored in dolavs (large meat crates) for a curing period of five days at 2°C to allow the brine to be absorbed and distributed within the back bacons. Vacuum packed backs were stored on top of each other in a dolav at nine levels. The positioning of every individual back in the dolav was recorded, which yielded in the further used class variable dolav level. After the five days curing period, the vacuum bags were removed and the backs were hung on racks for 60 minutes to remove unbound drip. After these 60 minutes, the backs were vacuum packed and weighed. The yield before frozen storage was calculated by using the fresh back bacon weight (before injection) and the back bacon weight after the curing period finished and after the backs were hanged on the racks for 60 minutes. After weighing, the backs were frozen for further processing.

$$\text{Yield before frozen storage} = \frac{W4}{W1} x100\%$$

W4 = weight of the back bacon after curing
W1 = weight of the back bacon before injection (fresh meat)

All backs were transported to a cold storage and frozen at a core temperature of -18 ˚C during 19 days. For experiment 1, sampling continued after the samples were frozen. For experiment 2, sampling was finished at this processing stage of back bacons. After 17 days of storage at -18 ˚C the frozen backs were transported during to a bacon slicing plant where the backs were tempered during two days to reach a core temperature of -8 ˚C. The tempered backs were sliced and packed into consumer packages (stacks) of eight slices of approximately 31 grams per slice. Two consumer packages were collected from the 10th vertebra from in total 274 random selected backs. These 274 samples were stored at 4 ˚C for 28 days (experiment 1). The consumer packages were opened after 28 days and the drip loss% during storage was determined and is referred to as yield consumer package. Yield consumer package was determined by measuring the total package weight, weight of the stack of bacon slices which were surface dried with a paper tissue and weighing the emptied package weight after it was dried with paper towels. The drip loss% during shelf life was expressed as yield consumer package:

$$\text{Yield consumer package} = 100 - \frac{W8 - (W7 + W6)}{W6} x100\%$$

W8 = Weight of the total consumer package
W7 = Weight empty and dried package
W6 = Weight bacon slices (surface dried with paper tissue) in consumer package

2.4 NIRS calibration and test-set (Experiment 1)

The NIRS dataset was split into two datasets; a calibration-set (70% of the samples) and a test-set (30% of the samples). The test-set samples were selected by OPUS 6.5 software (Bruker Optics GmbH, Ettlingen, Germany). Spectra data preprocessing and calibraton followed procedures which were described in Kapper et al. (2012). The measured spectral data were used for the prediction of the measured technological parameters like injection yield, curing yield, and yield consumer package on the data from the pre-selected samples in the test-set.

2.5 Statistical analysis

Meat quality measurements were evaluated by the mean, standard deviation, and the correlation coefficient (r). Outliers were removed ($\bar{u} \pm 2$ SD were considered as outliers). Statistical analysis was conducted using SAS® 9.1 (SAS Institute Inc., Cary, NC, USA) with level of significance set at $P < 0.05$. Pearson correlation coefficients were determined between pH, drip loss%, colour L*, injection yield, and yield consumer package using PROC CORR. Pearson correlation analysis was performed to evaluate the correlation between meat quality (pH, colour L*, drip loss%) and technological yield (injection yield, curing yield, yield consumer package).

For data of experiment 1 and 2, all curing yield values, yield consumer package values and yield before frozen storage values were corrected for dolav level and sample length. The injection yield values were corrected for the sample length only. The injection yield was calculated before the backs were placed in dolavs and are therefore not corrected for dolav level. For the data of experiment 1, the corrected values for curing yield, injection yield and yield consumer package were used for the NIRS prediction equations. In experiment 2, the corrected values for yield consumer package, injection yield and yield before frozen storage were used for the evaluation of the yield parameters between the pre-selected NIRS groups. For all GLM models of experiment 2, two-way interactions were tested between NIRS groups (high/low) and dolav level and no significant interaction was found in any of the used GLM models. NIRS spectra were analysed with chemometric data treatment by OPUS 6.5 software.

For experiment 1, the following GLM models were used:
Class: d (dolav level)

$$Y_{ijk} = \mu_i + d_j + l_k + e_{ijk}$$

Y_{ijk} is the dependent variable curing yield and Y_{ijk} is yield consumer package, μ_i is the mean value, d_j is the class variable dolav level, l_k is the sample length and e_{ijk} is the overall error.

The model for injection yield was without the class variable d (dolav level):

$$Y_{ij} = \mu_i + l_j + e_{ij}$$

For experiment 2, the following GLM models were used:
Class: d (dolav level) and NIRS selection (high/ low)
$Y_{ijkl} = \mu_i + d_j + \text{NIRS selection}_k + l_l + e_{ijkl}$

Y_{ijkl} is the dependent value curing yield and Y_{ijkl} is yield before frozen storage, μ_i is the mean value, d_j is the class variable dolav level, $high_k$ is the class variable from the NIRS selection (high or low), l_l is the sample length and e_{ijkl} is the overall error.

The model for injection yield was without the class variable d (dolav level):
Class: NIRS selection
$Y_{ijk} = \mu_i + \text{NIRS selection}_j + l_k + e_{ijk}$

3. Results and discussion

3.1 Experiment 1

The results from the meat quality and technological yield measurements of experiment 1 are shown in Tabel 1. The coefficient of variation (CV%) for pHu (3.6%), colour L* (9.9%) was comparable with previous results (pHu 2.1% - 3%, colour L* 7.3% - 10.2%) (Kapper et al., 2012). While the CV% for drip loss% (81%) was comparable with earlier results (53.7% - 79.7%) (Kapper et al., 2012). The low CV% values for pH, colour L* indicate that this dataset was made up with backs with limited variation on these parameters. The average drip loss% value (1.48%) was also lower than drip loss% values measured with the same method during a previous study (1.7% – 3.0%). Table 2 shows the Pearson correlation coefficients between the measured meat quality parameters (pHu, colour L*, and drip loss%) and the technological yield parameters (injection yield, curing yield, and yield consumer package) in experiment 1 correlations are low ($r \leq 0.26$), but most of them statistically significant ($P < 0.05$). Drip loss% did not correlate with injection yield (r -0.01, $P = 0.81$), which was not expected. Drip loss% and injection was regarded as related to WHC of the meat (Warriss & Down, 1985; Wismer-Pedersen, 1960). The pHu value positively correlated with curing yield and yield consumer package.

Table 1. Means, standard deviation (SD) of pHu, colour L*, drip loss%, injection yield, curing yield, yield consumer package, height, length, and width of bacon backs of experiment 1.

Parameter	Number	Mean	SD
pHu	351	5.54	0.2
Colour L*	351	55.6	5.5
Drip loss (%)	351	1.48	1.2
Injection yield (%)	345	116.4	2.1
Curing yield (%)	345	97.93	0.84
Yield consumer package (%)	274	99.78	0.10
Fresh deboned loin: Length (cm)	345	49.1	3.3
Width (cm)	345	19.2	0.7
Height (cm)	345	6.2	0.6

A higher ultimate pH results in higher curing yield and yield consumer package. Colour L* significantly correlated with injection yield and tended to be negatively correlated with curing yield and yield consumer package. Pork slices with higher drip loss% (lower WHC) had significantly lower curing yield and lower yield consumer package. The results agree with findings in literature (Fisher et al., 2000; Wismer-Pedersen, 1960) that meat with higher WHC, results in higher technological injections yields.

The data revealed that the back bacon length significantly affected the injection yield and therefore also the following processing yield parameters. Also the class variable dolav level was significantly affecting curing yield. The results showed that longer back bacon samples had significantly higher injection yield. Furthermore, the curing yield of back bacons was lower of samples that were stored at the lower levels in the dolavs. The curing yield on the samples from the bottom layer was 97.4% and the curing yield on the samples from the top layer was 98.6% (values were corrected by the GLM model for the sample length). The difference was significant ($P < 0.05$). Lower curing yields of samples stored at lower layers in the dolav might be caused by physical pressure on these samples, reducing the ability of these samples to retain the brine and drip, which reduce the correlation of WHC to injection yield.

Therefore it was chosen to correct the value of all samples for sample length. Curing yield and yield consumer package were also corrected for dolav level during storage (class variable). The yield values were corrected by the GLM procedure in SAS. The corrected curing yield and injection yield values were used for the calibration models with the NIRS spectra, to reduce the influence of factors on the yield values which did not depend on the meat quality (sample length and dolav level).

Table 2. Correlation matrix (Pearson correlation) of meat quality parameters, pH ultimate (pHu), colour L*, drip loss%, injection yield, curing yield, and yield consumer package of back bacon of experiment 1.

Parameter		pHu	Colour L*	Drip loss%
Injection yield	correlation (r)	0.08	-0.19	-0.01
	P value	0.13	< 0.001	0.81
Curing yield	correlation (r)	0.20	-0.10	-0.15
	P value	< 0.001	0.058	0.007
Yield consumer package	correlation (r)	0.18	-0.12	-0.26
	P value	0.003	0.054	< 0.0001

The aim of experiment 1 was to develop NIRS prediction equations for the injection yield, curing yield and yield consumer package values in bacon production. The developed prediction equations can be found in table 3. The prediction equation for pHu (R^2 0.26) was comparable with earlier results in Kapper et al. (2012), where no prediction equation for rough screening could be developed ($R^2 > 0.70$ and RPD > 1.9) for pHu values. The developed prediction equation for colour L* was within the range of results from the study of Kapper et al., 2012 (colour L*, R^2 0.23 – 0.69) and within the range of the review study of Prieto et al. (2009) (R^2 0.30 – 0.79). The developed prediction equations for drip loss% of a pork slice (R^2 0.50) was within the range of other studies for drip loss% (0.31 – 0.79) (Prieto et al., 2009). However, the developed prediction equation for drip loss% (R^2 0.50) was low in comparison to the results from Kapper et al., 2012 (drip loss%, R^2 0.54 – 0.76). The actual variation and the measured drip loss% values of the back bacons was low (average 1.48, range -0.52 – 5.47) and therefore it could be explained that the development of prediction equations (R^2 values) was somewhat reduced. The spectral resolution of the scans in experiment 1 was 32 cm^{-1}, while in Kapper et al. (2012) spectral resolution of 16 cm^{-1} was used. Similar protocols were used and the same device was used. The spectral data showed no obvious differences in absorption range and signal to noise between data from 32 cm^{-1} and 16 cm^{-1}. Thus we have chosen to reduce the spectral resolution to 32 cm^{-1} in stead of 16 cm^{-1}, which might also have reduced the development of accurate prediction equations, by reducing the sensitivity of the NIRS system to small absorption peaks.

Often a reduced spectral resolution improves the signal to noise ratio, when similar scanning time is taken. Thus, we have chosen to reduce the spectral resolution and keep the amount of scans similar which resulted in a reduced scanning time. The reduced scanning time together with the reduced spectral resolution might have reduced the ability of the development of prediction equations with higher R^2. However, this reduced scanning time had been chosen to perform measurements which are close to a practical application. The developed prediction equations for the technological meat quality (injection yield, curing yield, and yield consumer package) yielded in $R^2 < 0.21$ and RPD < 1.1 and showed very poor and not applicable prediction equations according to the scheme of Williams (2007), who indicated that an $R^2 \geq 0.70$ and RPD ≥ 1.9 are minimal requirements for NIRS application for rough screening.

Table 3. Calibration statistics on NIRS spectra obtained for the prediction of pHu, colour L*, drip loss%, injection yield, curing yield, and yield consumer package on Longissimus dorsi samples for bacon production, by using a calibration and test-set validation of experiment 1.

Parameter	Spectra treatment	Calibration				Test-set validation				
		n	R^2	RMSEE	RPD	n	R^2	RMSEP	RPD	Bias
pHu	A	241	0.16	0.18	1.09	103	0.26	0.18	1.16	-0.01
Colour L*	A	241	0.55	3.68	1.49	103	0.53	3.93	1.47	0.62
Drip loss (%)	A	241	0.55	0.78	1.48	103	0.50	0.91	1.47	0.25
Injection yield (corrected by GLM)	C & B	241	0.18	0.40	1.10	103	0.23	0.45	1.15	-0.05
Curing yield (%) (corrected by GLM)	C & B	241	0.04	0.35	1.02	103	0.08	0.39	1.04	-0.02
Yield consumer package (%) (corrected by GLM)	C & D	192	0.06	0.10	1.03	82	0.08	0.10	1.04	0.00

Treatment A is no spectral data pre-processing, treatment B is straight line subtraction, treatment C is 1^s derivative with 17 data point smoothing, treatment D multiplicative scatter correction.

3.2 Experiment 2

Sorting pork meat samples for WHC using NIRS had significant effect on pHu and colour L*. The high NIRS drip loss% group resulted in significantly ($P < 0.05$) lower pHu value (5.50 vs 5.58) and higher colour L* (56.9 vs 53.8) compared to the low NIRS group. The differences between the groups show that the pHu and the colour L* of the predicted drip loss% groups were as expected for meat with a higher WHC (lower drip loss%) (Eikelenboom et al., 1974; Fisher et al., 2000).The drip loss% groups were pre-selected by the NIRS prediction equation, developed in Kapper et al. (2012). The values in table 4 show the technological yield parameters (injection yield, curing yield, and yield before frozen storage) and show that the differences of the values between the low and high predicted groups are very small. There was no significant difference for injection yield, curing yield and yield before frozen storage between the low and high predicted drip loss% groups.

Table 4. Mean and standard deviation (SD) of injection yield, curing yield, and yield before frozen storage of loin samples categorized into two quality groups (low and high predicted drip loss%) on the basis of predicted WHC of each sample by NIRS technology in experiment 2.

	Low predicted drip loss (< 1.47%)		High predicted drip loss (> 3.14%)	
	Mean	SD	Mean	SD
Number of samples	128		152	
Injection yield (%)	116.4	1.8	116.5	1.7
Curing yield (%)	98.7	0.6	98.6	0.5
Yield before frozen storage (%)	114.8	1.7	114.8	1.7

All measured parameters were not significant different (P < 0.05).

The corrected values for the covariables sample length and dolav level are expressed in table 5. The R^2 value of the GLM models in SAS showed that less than 10% of the variation was explained ($R^2 < 0.10$). The low R^2 values in de GLM models indicated that there was no effect of pre-selection on fresh meat quality on the technological yield values. The values in table 5 show that injection yield and curing yield are not significant different between the pre-selected groups (low/high) after correction for sample length and dolav level. The low drip loss% group had no significant different, injection yield (16.8%) in comparison to the high drip loss% group (16.9%). However the injection yield was not significant different, lower injection yield values of meat with higher WHC was also observed in earlier unpublished results. It is considered that the more intact and more firm meat is more resistant against the uptake of the brine secretion by the injector needles.

Table 5. Means and standard deviations (SD) of the technological yield corrected for sample length and dolav level of loin samples categorized into two quality groups (low and high drip loss%) on the basis of predicted WHC by NIRS technology.

	Low predicted drip loss (< 1.47%)		High predicted drip loss (> 3.14%)	
	Mean	SD	Mean	SD
Number of samples	128		152	
Injection yield (%)	116.8	0.3	116.9	0.4
Curing yield (%)	98.7	0.2	98.6	0.1
Yield before frozen storage (%)	84.8	0.3	84.7	0.4

All measured parameters were not significant different

The pre-selected high drip loss% group showed no significant lower yield before frozen storage in comparison with the low drip loss% group. It is observed that sorting for fresh meat quality with NIRS results in significant differences of the average fresh meat quality parameters pHu (5.58 versus 5.50) and colour L* (53.8 versus 56.9) in the pre-selected groups. Thus it is considered that unaccountable factors and processing variation reduce the significant difference between both WHC quality groups to such extent that no relevant yield differences could be observed. No differences were observed in drip loss% during the curing process (curing yield) of the bacon backs. Thus, the difference between the curing yield values tended ($P = 0.07$) to be higher for the high WHC group. The relative short period of curing period of five days might have been too short to show a significant lower brine loss for the low drip loss% group. Also Warriss and Down (1985) observed lower injection yield values and higher curing yield values in meat which was regarded as having higher water holding capacity (48 hours fasting of the pigs) than in meat which was regarded as having lower WHC (4 hours fasting of the pigs) (Warriss & Down, 1985)

4. Conclusions

In this study it was investigated if NIRS prediction equations are relevant for technological yields for bacon production. Results showed that measurements on fresh meat primals, such as pHu, colour L*, and drip loss% significantly correlate with measured yield parameters in back bacon production ($r \leq 0.26$). However the observed correlations were small. When the WHC of fresh backs was measured with NIRS and sorted on the predicted WHC, no significant differences could be observed between the high WHC group and the low WHC group for the technological yields (injection yield, curing yield, and yield before frozen storage), however, significant differences were found on pHu and colour L* between both pre-selected NIRS grouped backs. Results showed that NIRS could not be used to optimize the production yield for back bacon production and are considered of no relevance for the production of back bacon.

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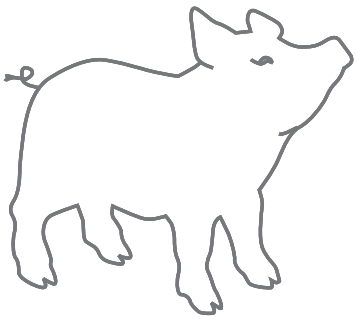

Chapter 5

Using near infrared spectroscopy (NIRS) and pork quality measurements to correlate and predict yields during cooked ham production

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Abstract

The potential of fresh pork quality measurements and near infrared spectroscopy (NIRS) water holding capacity (WHC) to correlate and predict cooked ham production yields was investigated. Fresh meat quality (pHu, L* value, drip loss%) were measured on loins and hams from 160 random pigs. Cooked ham yields (transport loss, injection, and tumbling yield) were determined on the individual or combined silverside and topside muscles (cooking and final yield). Ultimate pH from the ham correlated with all yield values (r = -0.19 to r = -0.55). NIRS prediction on the ham had highest correlation for cooking and final yield (r = -0.42 and 0.37). Cooking yield correlated significantly with colour L* (r = -0.20) and pHu value of the loin (r = 0.23). Transport loss of ham muscles correlated significantly with cooking (r = -0.36) and final yield (r = 0.35), while drip loss% of loin samples also correlated significantly with cooking (r = -0.32) and final yield (r = 0.36). Pre-selection of hams by NIRS into three predicted WHC (drip loss%) groups resulted in differences between the low and high WHC groups for cooking yield and final yield. It was concluded that NIRS can be used to predict fresh ham quality for sorting and optimization of the cooked ham process.

Key Words: NIRS, drip loss%, water holding capacity, ultimate pH, colour, pork quality, cooking yields

1. Introduction

Water holding capacity (WHC) is one of the most important pork quality traits as it affects the sensory appreciation of pork by consumers, determines amount of saleable meat, and increases processing yield of further processed products (Andres et al., 2007; Bowker et al., 1999; Cassens et al., 1975; Channon et al., 2002; Rosenvold & Andersen, 2003). The pork processing industry has been specialized in sorting on weight and lean characteristics of carcasses and primal cuts (Kapper et al., 2012). However, sorting for WHC by rapid on-line non-invasive pork quality measurements has not been achieved yet. Near infrared spectroscopy (NIRS) was identified as a potential measuring technique to predict WHC of pork primals (Barlocco et al., 2006; Geesink et al., 2003; Kapper et al., 2012; Prieto et al., 2009; Swatland & Irie, 1992). NIRS could potentially also be used for sorting primal cuts according to different WHC categories (Barlocco et al., 2006; Kapper et al., 2012; Prieto et al., 2009; Swatland & Irie, 1992). Prediction and sorting of the pork primals according to their WHC might improve processing yields and reduce sensory variation in cooked products. Swatland and Barbut (1995) showed that myofibrillar NIR birefringence in turkey meat correlated with WHC of the fresh meat and correlated with the fluid loss during cooking (Swatland & Barbut, 1995). Kapper et al. (2012) concluded that NIRS could be used to select for fresh pork WHC (24 hours post mortem) for different market segments or processing applications under commercial cold room conditions. The objective of this study was to determine the fresh pork quality measurements and NIRS WHC prediction of ham primals to optimize cooked ham production yields.

2. Materials and methods

2.1 Muscle collection and pork quality measurements at a pig processing plant

A total of 160 pig carcasses were randomly chosen at one pig processing plant. The pigs were electrically stunned and had a carcass weight of approximately 92 kg. At 24 hours post mortem one middle and ham primal were labelled and used for the experiment from the right side of these 160 carcasses. Ultimate pH (pHu) (MPI pH-Meter, Meat Probes, Inc), colour L* (Chroma Meter CR-400, Konica Minolta Sensing, Inc.), and 48 hours drip loss% of the loin muscles samples were measured according to (Kapper et al., 2012). Colour L* and pHu was measured in duplo and averaged. A slice of approximately two cm thick was taken from the *Longissimus dorsi* (loin) at the shoulder side for the determination of drip loss%. During 48 hours at 4°C in consumer retail trays. Colour L* and NIRS measurements were taken on the fresh cut surface of the ham primal, which was the area where it was separated from the middle between the second and third vertebra in the *Lumborum* area. The fresh cut surfaces of the ham primals were bloomed for more than 30 minutes, before sampling was performed. A NIRS scan of the fresh cut surface of the ham primal was also subjected to an earlier developed prediction model from Kapper et al. (2012) to predict drip loss%.

The NIRS scans of the fresh cut surface of the ham primals were used to develop a prediction equation for cooking yield measured on the actual cooked hams. Spectra were measured with a Bruker Matrix-FE (Bruker Optics GmbH, Ettlingen, Germany), using a contact free probe at a distance of approximately 12 centimetres from the sample surface (Q-412 Bruker Optics GmbH, Ettlingen, Germany) with an integrated light source in the probe. The device measured reflectance from 4.000 to 12.000 cm^{-1} and 32 scans were averaged per sample. The chosen wavelength resolution was 16 cm^{-1}. The total scan time was approximately 10 seconds per sample. The NIRS spectrum was automatically transformed into Log 1/R absorption spectrum by the software and saved for further analysis (OPUS 6.5 software, Bruker Optics GmbH, Ettlingen, Germany).

All ham primals were individually labelled and identified for the duration of the experiment. After the fresh meat measurements, the ham primals were deboned and the silverside and topside muscle from every ham primal were placed in meat crates attached to their individual label. Colour L* and pHu of the silverside and topside muscle was determined and all muscles were weighed. Labels were attached to the muscles with a plastic cord and all samples were collected in one dolav for next day's transport to a ham processing plant.

2.2 Cooked ham processing and yield measurements

At 72 hours post mortem, the silverside and topside muscles arrived at the cooked ham processing plant. After arrival all muscles were weighed to determine their weight loss% (transport loss) during transport. All muscles were injected with brine directly after weighing. The amount of injected brine was set to approximately 15% of the initial muscle weight. The injection was set to reach 0.78 g/kg, E450, stabilizer, 0.70 g/kg, E301, antioxidant, and 1.8% Nitrate Phosphate salts (NPS) in the cooked ham end product. Components in the brine were: water (H_2O), NPS (nitrate containing salts): NaCl (Sodium Chloride), $NaNO_2$ (Sodium Nitrite E250), antioxidants $C_6H_7O_6Na$ (Sodium Ascorbate E301), and functional phosphates P_2O_7 (diphosphate E450). The injection device operated with vertical moving needles injecting the individually labelled muscles. All muscles were weighed immediately after brine injection, after which they were simultaneously tumbled under vacuum at approximately 1˚C for five hours. About 3.3% of the total meat weight before injection was added as brine during tumbling.

After tumbling, the muscles were weighed and both silverside and topside muscle of the same ham were placed together in one traditional cooking can with a pressurised lit. Muscles were placed in open plastic bags in the cans to allow for moisture release from the muscles during cooking. The cans were placed on trailers which were placed in a cooking unit using 75˚C during the cooking process, which continued until a core temperature of 70˚C was reached. After cooking, the canned hams were showered with water of approximately 10˚C and the cooked hams were cooled to 4˚C during one day. The day after cooking all hams were taken out of the cans, vacuum packed, and stored at 4˚C. Two days after vacuum packaging, the plastic bags were removed and the cooked hams were weighed to be able to determine weight loss during cooking (cooking yield %).

2.3 Statistical analysis

Statistical analysis was conducted using SAS® 9.1 (SAS Institute Inc., Cary, NC, USA) with level of significance set at $P < 0.05$. Data were analyzed using PROC GLM and PROC CORR (Pearson correlation). PROC GLM was used to evaluate significant relations between the fresh meat quality and the technological quality parameters of hams grouped by the predicted drip loss% values and by the predicted cooking yield values. Finally, Pearson correlation coefficients were determined between pHu, drip loss%, colour L*, injection yield, tumbling yield, and cooking yield using PROC CORR. The following calculations were used:

Transport loss = ((weight after transport – weight before transport)/ weight before transport)*100%
Injection yield = (weight after injection / weight before injection)*100%
Tumbling yield = (weight after tumbling / weight before tumbling)*100%
Cooking yield = (weight after cooking / weight before cooking)*100%
Final yield = (weight fresh silverside + weight topside muscle after deboning / weight cooked ham)*100%

NIRS spectra from the fresh cut surface of the ham primals were analysed with chemometric data treatment by OPUS 6.5 software (Bruker Optics GmbH, Ettlingen, Germany). The NIRS spectra taken from the fresh cut surface of the ham primals were used for the prediction of drip loss%. The NIRS spectra from the fresh cut surface of the ham primals were also used to develop a prediction equation for the cooking yield values. Modified partial least square regression (MPLS) was used for the development of the prediction equation for cooking yield. Different spectral data pre-treatments were investigated using 1st or 2nd derivative with smoothing, vector normalization, constant offset elimination and no spectral data pre-processing. Also combinations of data pre-treatments were investigated. The chemometric prediction equation was set to a maximum rank of 10. Seventy % of the collected NIRS spectra were used for the development of the prediction equation and 30% of the samples were used for the evaluation of the prediction equation (test-set). The test-set samples were pre-selected by the OPUS software which selected the samples in such a way that the test-set and the calibration set had both almost similar variation and range.

The results from the developed prediction equation were evaluated by the R^2, Root Mean Square Error of Prediction (RMSEP), and the Relative Prediction Deviation (RPD) of the test-set. The predicted drip loss% values from the NIRS spectra of the fresh cut surface of the ham primals was used to group the cooked hams in three quality groups: high, moderate, and low. The grouped hams were selected by taking arbitrarily the lowest 20% of predicted drip loss% values as the high WHC groups and the highest 20% of the predicted drip loss% values as the low WHC group. The 60% of the ham primals which did not belong to the high or low WHC group were classed as moderate WHC samples.

3. Results and discussion

3.1 Evaluation of meat quality measurements

Means and standard deviation of the meat quality measurements of the loin, the ham primal (cut surface) and of two individual ham muscles are displayed in table 1. The ham muscles differed significantly for the meat quality values (pHu, L*). There was no significant difference in transport loss between the silverside and topside muscles. The loin drip loss% values had a coefficient variation (CV) of 61%, which indicates that they had a lower variation than an earlier study (CV = 80%, (Kapper et al., 2012). Kapper et al. (2012) showed considerably higher CV values (15 – 27%) for drip loss% than found by Savenije et al. (2006).

Table 1. Means and standard deviations (SD) of meat quality measurements (L*, pHu, drip loss%, predicted drip loss% by NIRS, transport loss%) of the loin samples and, the fresh cut surface of the ham primals, and of the silverside and topside muscles.

	pHu		L*		Drip loss%		NIRS predicted drip loss%		Transport loss%	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Loin muscle	5.56^{a}	0.13	48.9^{a}	3.9	1.7	1.1				
Fresh cut surface ham primal	5.63^{bc}	0.20	50.2^{b}	4.0			0.9	1.8		
Silverside muscle	5.66^{b}	0.23	58.0^{c}	3.6					1.6^{a}	1.0
Topside muscle	5.60^{ac}	0.19	54.0^{d}	3.3					2.2^{a}	1.1

^{a,b,c,d} letters indicate significant difference (P<0.05) between the mean values of each column.

3.2 Correlations between the fresh meat quality measurements

Pearson correlations between the fresh meat quality parameters of the loin, fresh cut surface of the ham primal, and both deboned ham muscles (silverside and topside muscle) are showed in table 2. The pHu of the loin muscle correlated significantly with its drip loss% (r = -0.18, $P < 0.05$). The colour L* of the loin also correlated significantly with its drip loss% (r = 0.47, $P < 0.001$). These results are in agreement with results from other researchers (Eikelenboom et al., 1974; Fischer, 2007; Huff-Lonergan et al., 2002).

3.3 Correlations between meat quality measurements and carcass locations

All measured pHu values correlated consistently to meat quality parameters (pHu, L*, drip loss%, transport loss), of the loin, the fresh cut surface of the ham primal, and of the deboned ham muscles (Table 2). Colour L* of the loin had similar correlations with the measurements at other muscles/locations. The results from table 2 indicate that meat quality measurements carried out at one carcass location (loin) can be used to estimate the overall ham meat quality, despite separate muscles differing significantly within a carcass (Table1).

3.4 Correlations between meat quality measurements measured at the ham and processing yields

The injection and tumbling yields of the individual silverside and topside muscles are shown in table 3 and were significantly different between both muscles ($P < 0.001$). In table 4 the correlations between meat quality measurements the ham primal (pHu, L*, NIRS predicted drip loss%, transport loss values of deboned ham muscles) and processing yield values (injection, tumbling, cooking, and final yield) are shown. The ham pHu (r -0.19 - -0.55) and the transport loss values (r 0.29 – 0.38) correlated significantly ($P < 0.05$) with all processing yield values. Colour L* value of the ham primals did not correlate significantly with the final yield and revealed lower correlations than the ultimate pH values. The transport weight loss is a direct indication of WHC and also showed significant correlations with cooking (r = -0.36, $P < 0.001$) and final yield (r = 0.35, $P < 0.001$) (Table 4). The NIRS values from the ham primals did not correlate significantly with injection yield (r = 0.02) and showed a lower correlation for tumbling yield (r = 0.27) than the correlations of pHu, L*, and transport loss (r = 0.17 - -0.55) to tumbling yield. However, NIRS values of the ham primals had highest correlations with cooking and final yield (r = -0.42 and 0.37, $P < 0.001$).

Table 2. Pearson correlation coefficients between fresh meat quality parameters (L*, pHu, drip loss%, predicted drip loss% by NIRS, transport loss%) of the loin, fresh cut surface of the ham primal, and the silverside and topside muscles.

		Loin muscle		Fresh cut surface ham primal			Silverside muscle			Topside muscle		
		L*	Drip loss%	pHu	L*	NIRS predicted drip loss%	pHu	L*	Transport loss (%)	pHu	L*	Transport loss (%)
Loin Muscle	pHu	-0.19*	-0.18*	0.47**	-0.39**	-0.26**	0.42**	-0.28**	-0.32**	0.51**	-0.44**	-0.44**
	L*	1.00	0.47**	-0.50**	0.35**	0.14*	-0.50**	0.30**	0.39**	-0.49**	0.39**	0.33**
	Drip loss%		1.00	-0.20*	0.12	0.25*	-0.20*	0.07	0.39**	-0.19*	0.27**	0.37**
Fresh cut surface ham primal	pHu			1.00	-0.46**	-0.27**	0.98**	-0.48**	-0.61**	0.98**	-0.54**	-0.55**
	L*				1.00	0.44**	-0.43**	0.40**	0.29**	-0.48**	0.50**	0.41**
	NIRS predicted drip loss%					1.00	-0.23*	0.06	0.36**	-0.30**	0.22*	0.39**
Silverside muscle	pHu						1.00	-0.48**	-0.61**	0.92**	-0.53**	-0.52**
	L*							1.00	0.17*	-0.46**	0.48**	0.16*
	Transportloss (%)								1.00	-0.59**	0.34**	0.75**
Topside muscle	pHu									1.00	-0.52**	-0.56**
	L*										1.00	0.48**

** indicates significant correlation at the P<0.05 level and ** indicates significant correlation at the P<0.001 level.*

3.5 Correlations between loin meat quality measurements and ham processing yields

Analysis revealed that cooking yield of the hams significantly correlated with the colour L* (r = -0.20, $P < 0.05$) and the pHu value (r = 0.23, $P < 0.05$) of the loins. Similar significant correlations were observed between the loin drip loss% values and both cooking (r = -0.32, $P < 0.001$) and final yield (r = 0.36, $P < 0.001$). These results are not in accordance with Bertram et al. (2003), who did not found significant correlations between cooking yield and pHu and drip loss% (using the Honikel method).

Table 3. Means and standard deviations (SD) of the injection and tumbling yields from the silverside and topside muscles, and cooking and final yield of the combined silverside and topside ham muscles.

	Silverside muscle		Topside muscle		Silverside and topside together	
	Mean	SD	Mean	SD	Mean	SD
Injection yield (%)	114.81[a]	1.71	116.55[b]	1.92		
Tumbling yield (%)	102.78[a]	1.23	103.85[b]	1.58		
Cooking yield (%)					88.33	2.56
Final yield (%)					103.27	3.40

[a,b]letters indicate significant difference (P<0.001) between the mean values of each row

3.6 Comparing NIRS and fresh meat quality measurements for applicability

Drip loss% after 48 hours storage of loin samples or transport loss values (24 hours storage) of ham muscles can be used to significantly estimate both cooking and final yield. However, the drip- and transport loss measurements are time consuming and not practical. A less time consuming, but still an invasive measurement, is the pHu assessment, which correlated significantly with processing yields of the ham muscles and has also been suggested by other researchers (Cariou et al., 1988; Eikelenboom et al., 1974; Flores et al., 1999). The NIRS measurements are fast (~10 sec), non invasive and revealed significant correlations with processing yields (cooking and final yield), and showed slightly higher correlations for both the cooking (-0.42, $P < 0.001$) and final yield (0.37, $P < 0.001$) than the pHu correlations (0.38, $P < 0.001$ and -0.19, $P < 0.05$). We conclude therefore that NIRS can be a fast and non-invasive technology to predict processing yields of ham muscles and revealed in this experiment the best option for a practical application.

Table 4. Correlations between ham meat quality parameters (L*, pHu, predicted drip loss% by NIRS, and transport loss) measured at the fresh cut surface of the ham primal and the processing yield values (injection yield, tumbling yield, cooking yield, and final yield).

Processing yield values (silverside and topside muscle together)	Fresh cut surface ham primal pHu	L*	Predicted drip loss (%) (NIRS)	Transport loss
Injection yield (%)	-0.32**	0.17*	0.02	0.29**
Tumbling yield (%)	-0.55**	0.32**	0.27**	0.38**
Cooking yield (%)	0.38**	-0.22*	-0.42**	-0.36**
Final yield (%)	-0.19*	0.12	0.37**	0.35**

** indicates significant correlation at the P<0.05 level and ** indicates significant correlation at the P<0.001 level.*

3.7 Grouping ham primals by NIRS measurements

The NIRS prediction values from the fresh cut surface of the ham primals were evaluated by categorizing the hams into three groups depending on their predicted drip loss% value. Table 5 shows the cooking and final yield values of the hams categorized into three quality groups (Low, Moderate, and High) depending on the predicted drip loss% value of the ham primal by NIRS technology. The 20% lowest and 20% highest predicted drip loss% values by NIRS were arbitrary selected as a high WHC and a low WHC group, respectively. The low WHC group had a significantly ($P < 0.05$) lower cooking and final yield than the moderate and high WHC group. The moderate WHC group tended to have a lower cooking yield than the high WHC group but this was not significant ($P = 0.052$). Post mortem degradation of desmin (a cytoskeletal protein) could explain the difference in cooking yields between the WHC groups. It is expected that more desmin was released in low WHC muscles and created cavities in the meat, which might have enhanced exudation during cooking for the low WHC groups (Huff-Lonergan & Lonergan, 2005; Melody et al., 2004). Furthermore, post mortem accumulation of the hydrogen ions might be related to the forming of drip channels in the muscle by aggregation and denaturation of muscle proteins (Honikel et al., 1986; Lawson, 2004). The moderate WHC group had a significant ($P = 0.01$) lower ultimate pH (5.62) than the high WHC group (5.74), which is in accordance with this theory. The low WHC group was not significantly different in pHu (5.54) from the moderate WHC group ($P = 0.108$).

3.8 Prediction equation by NIRS for cooking yield

The NIRS spectra from the fresh cut surface of the ham primals and the cooking yield values were used to develop a prediction equation for cooking yield. The prediction equation revealed R^2 0.5, RMSEP 2.2, RPD 1.5 of the test-set samples, which is considered as not optimal for rough

screening (Kapper et al., 2012; Williams, 2007). The results reveal that the NIRS prediction of cooking yield or NIRS prediction of drip loss% might be useful for pre-selection of ham primals for optimization of the cooking process, but that the prediction equations still need further improvements ($R^2 > 0.70$, RPD > 2.0) (Kapper et al., 2012; Williams, 2007).

Table 5. Mean and standard deviation (SD) of the cooking yield of hams categorized into three quality groups (Low, Moderate, and High) on the basis of the 20% lowest predicted drip loss% values (High WHC groups), 20% highest predicted drip loss% values (Low WHC group) and 60% of the moderate predicted drip loss% values (Moderate WHC group) by NIRS on the fresh cut surface of ham primals.

	High WHC		Moderate WHC		Low WHC	
	Mean	SD	Mean	SD	Mean	SD
Number of samples	32		96		32	
Cooking yield%	89.66 [b]	1.98	88.50 [b]	2.28	86.48[a]	2.89
Final yield%	104.93 [b]	2.87	103.82 [b]	3.30	101.14[a]	4.03

[a,b]letters indicate significant difference (P<0.05) between the mean values of each row

3.9 Phosphate level of the hams

Phosphate has a positive contribution to the properties and water holding capacity of meat and is therefore often added to cooked products (Cassidy et al., 1978). The used level of 0.78 g/kg NPS is relatively low in comparison to current commercial available cooked hams, which may contain maximum 3.0 g/ kg P_2O_5 (BMPA, 2010). We assumed that the yield differences between the pre-selected NIRS groups might have been smaller when more phosphates were added to the hams due to the expected water binding properties of phosphates.

4. Conclusions

WHC and pHu measurements correlated significantly with injection, tumbling, cooking, and final yields. WHC and pHu measurements are regarded as time consuming, invasive and less practical to estimate ham processing yields. NIRS predictions of WHC (drip loss%) on the fresh cut surface of ham primals showed significant correlations for cooking and final yield, were non-invasive, and fast (~10 sec). It was therefore concluded that the NIRS measurement were more applicable, even though prediction equations to estimate cooking yields can be further improved ($R^2 > 0.70$, RPD > 2.0). Pre-selection of fresh hams by NIRS predicted WHC (drip loss%) resulted in significant differences between the low WHC group and the moderate or high WHC group for the cooking and final yield values. It was concluded that NIRS can be used to predict fresh ham quality for sorting and optimization of the cooked ham process.

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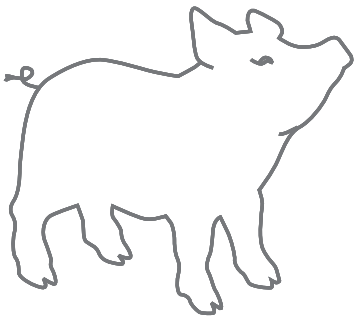

General discussion

1. General discussion

This thesis emphasizes the importance of rapid prediction of pork water holding capacity (WHC), which is regarded as one of the most important pork quality aspects related to consumer perceptions and processing yields (Briskey et al., 1960; Eikelenboom et al., 1974; Melody et al., 2004; Nelson, 2005; Offer, 1988a; Puolanne & Halonen, 2010; Rosenvold & Andersen, 2003; von Rohr, 1999; Warriss & Down, 1985; Wismer-Pedersen, 1960). Factors influencing pork WHC are fairly well understood, however limitations in current WHC prediction techniques limit rapid and accurate predictions of pork WHC post mortem. This thesis consists of two objectives. The first objective was to investigate the possibilities of prediction of pork WHC by measuring parameters such as pH, colour L*, drip loss%, water absorption, and by measuring near infrared spectroscopy (NIRS) spectra (chapter 1, 2, and 3). The second objective was to investigate if predictions of WHC could be used to optimize pork processing yields (chapter 4 and 5).

2. Variation pork quality

Pork water holding capacity (WHC) affected the amount of saleable meat through loss of product as a result of drip loss% (chapter 1, and 2, and 3) and affected processing yields of further processed products (chapter 4, and 5) (Cariou et al., 1988; Flores et al., 1999). Scientific knowledge of perimortem muscle metabolism is applied to reduce and control factors that affect this metabolism, and focus on genetics, fasting strategies, slaughter practices, and carcass cooling procedures (Briskey & Wismer-Pedersen, 1961; Channon et al., 2002; Dransfield & Lockyer, 1985; Enfalt et al., 1993; Jones et al., 1993). Eventhough, perimortem muscle metabolism is largely understood, pork WHC varies between animals and pig processing plants (chapter 2). In chapter 2 it was observed that mean drip loss% values differed significantly between four pig processing plants A (2.1% ±1.7), B (1.7% ±1.2), C (2.7% ± 1.5), and D (3.0% ± 1.8), $P< 0.001$. We expect that differences between pig processing plants are confounded with factors such as genetics, animal handling, slaughter procedures, and cooling practices. Hambrecht (2004) revealed that pig processing plants can affect pork WHC by their management, but that the variation in WHC cannot be controlled completely. Pork WHC will always vary between carcasses. For example, pig processing plants can affect the speed of post mortem metabolism of carcasses by the rate of carcass cooling (Hambrecht, 2004; Maribo et al., 1998). Increased temperature decline of a pork carcass will reduce post mortem muscle metabolism and improve pork WHC. Nevertheless, carcasses vary in weight (74 kg through 112 kg) and muscle thickness (46 mm through 79 mm), which provokes variation in temperature decline and is one of the reasons of variation in WHC. Results from chapter 2 revealed that WHC in pig processing plants had a coefficient of variation (CV) of 54% to 80%. These results are in agreement with Hambrecht (2004). Nowadays, pork is sorted and validated on weight and lean characteristics at pig processing plants, while sorting for WHC is not applied. Therefore, all currently sorted carcasses and primals at pig processing plants vary largely in WHC.

3. Prediction and correlation to WHC

3.1 pH

Undoubtedly, pH is the parameter which is most often used to predict pork WHC at pig processing plants as it reveals post mortem lactic acid accumulation, which affects the development of WHC (Cariou et al., 1988; Eikelenboom et al., 1974; Kauffman, 1993; Van Oeckel et al., 1999). Antemortem metabolism and animal physiology affect the rate and extent of pH decline in muscle and pork (Sellier & Monin, 1994). Results in chapter 1, 2, and 5 showed that pH values had significant but low correlations with WHC ($r = 0.18$ to $r = 0.53$, $P < 0.05$). Literature suggests that post mortem processes such as muscle protein denaturation and aggregation, enzymatic cytoskeletal protein catabolism, the intensity of rigor, and the level of drip channels formation, affect WHC development (Huff-Lonergan & Lonergan, 2005; Lawson, 2004; Nelson, 2005; Schäfer et al., 2002). These post mortem processes are only partly influenced by the rate of pH decline and ultimate pH of meat, which decreases the correlation of pH with WHC. The correlation between WHC and pH in chapter 5 is in accordance with literature ($r = 0.28 – 0.33$, $P < 0.01$) (Eikelenboom et al., 1974; Huff-Lonergan et al., 2002). In addition, pH measurements on meat cannot be performed contact free. Insertion of a glass pH probe is needed to measure pH values in meat, whereby chemical and physical residues can be a complicating factor for food safety. Measurements of pH to predict pork WHC were therefore regarded as less applicable for an accurate prediction of WHC.

3.2 Colour

Colour measurements of meat (L*, a*, and b* values) relate to pork WHC. In general, pork with higher WHC has a darker colour (Huff-Lonergan et al., 2002; Mancini & Hunt, 2005). Correlation coefficients (r) between colour L* and WHC values were found to range from -0.32 through -0.68 ($P < 0.05$) in chapter 1, 2, and 5. In literature correlations can be found between colour L* and WHC ($r = -0.33$ through -0.44, $P < 0.001$) (Huff-Lonergan et al., 2002). Colour values are used to measure myoglobin concentration and the degree of oxygenation of myoglobin in pork (Beriain et al., 2009). Oxidation of myoglobin (purple colour) into oxymyoglobin (red cherry colour) and finally into metmyoglobin (brown colour) is the result of oxidation of both ferrous myoglobin derivatives (Brewer et al., 2001; Mancini & Hunt, 2005). A colour value measured on pork is affected by the amount of (oxidized) myoglobin, and the amount of light scatter (reflection) from water present on the surface. Pork with low WHC has more denatured proteins, which increases scattering and dispersion of light on the meat surface, resulting in increased lightness and paleness of pork (Beriain et al., 2009). The concentration and extent of oxidation of myoglobin is poorly related to pork WHC. However, myoglobin affects the measured colour value and thereby reduces the accuracy of a colour measurement to predict WHC (Beriain et al., 2009). Colour measurements were therefore regarded as less applicable for accurate prediction of pork WHC.

3.3 Drip loss

Drip loss% is the exudation of moisture from meat, and is a direct measurement of WHC. However, drip loss% measurements require preparation of pork samples (chapter 3). In addition, drip loss% measurements are time consuming, and need a minimal time of 60 minutes (centrifugation) or 24 hours to 48 hours (hanging bag, EZ – cup, case-ready tray) before the results are available (chapter 3) (Bertram et al., 2001; Honikel, 1998). Drip loss% measurements were regarded as not practical for a rapid indication of WHC because they are costly, invasive, and require extensive preparation and sampling time.

3.4 Early post mortem water absorption

In chapter 3 it was investigated if the quantity of unbound, or less bound water, in meat ran parallel with WHC of pork, and if measurements of unbound water early post mortem could be used for prediction of WHC (Fischer, 2007; Honikel, 1998; Puolanne & Halonen, 2010). Chapter 3 showed that early post mortem moisture absorption (15 min insertion time) by cotton-rayon material correlated significantly with drip loss% measured at 24 hours post mortem ($r = 0.33$ through 0.48, $P < 0.05$). Individual absorption data revealed that time frames of 75 minutes through 120 minutes, 240 minutes through 270 minutes post mortem correlated significantly to WHC. However, correlations to WHC ranged from r -0.33 through -0.48, $P < 0.05$) during the first five hours post mortem and were considered as relatively low in comparison to NIRS WHC predictions ($R^2 = 0.54$ through 0.76) in chapter 2. These correlations were within the range of correlations of pH and colour with WHC, and varied greatly (chapter 1, 2, and 5; $r = 0.19$ through 0.53). The early post mortem absorption method was invasive, required extensive handling during sampling, and needed 15 minutes dwell time (chapter 3). Practical implications of this method were expected to be limited. The method might be evolved by further research into a system measuring rapid (< 15 minutes) water absorption in carcasses early post mortem with a potential for rapid prediction and practical application.

4. Near infrared spectroscopy (NIRS)

Chapter 1 revealed that NIRS could be used for rough screening of pork for WHC (prediction equation, $R^2 = 0.73$) from data of pig processing plant A. However, the residual prediction deviation (RPD) was still low (≤ 2.0). In chapter 2 it was concluded that NIRS can be seen as a technique suitable for non invasive prediction of WHC under pig processing plant conditions ($R^2 = 0.54$ through 0.76) (Kapper et al., 2012; Prieto et al., 2009). NIRS prediction of WHC was rapid (< 10 sec) and had a acceptable coefficient of determination ($R^2 = 0.73$) with WHC measured and was therefore regarded as suitable for rapid prediction of pork WHC under commercial production conditions.

4.1 Developed NIRS prediction equations

The prediction equations for WHC in chapter 1, 2, 4, and 5 revealed that best predictions were gained without spectral pre-processing before partial least squares (PLS) calibrations of NIRS data. However, we considered that it is favourable to apply a pre-treatment on NIRS spectra, such as a derivative or vector normalization to reduce baseline differences. Baseline differences that are caused by non WHC related factors reduce correlations of the NIRS spectra with WHC and should be minimized. For example, the light source of the NIRS device is a non WHC related factor, and can vary by changes in the environment, and therefore vary in intensity during the measurement, which causes the spectra baseline to shift. In addition, meat continuously changes over time, which can affect the NIRS measurement. For example, accumulation of expelled water (drip loss%) on the fresh cut surface of a meat sample might become more pronounced over time, which can induce light scatter and baseline shifts during NIRS measurements. These possible sources of external variation show the inmportance of standardisation of the sampling procedure. The prediction equation for drip loss% in chapter 1 (no pre-processing: R^2 0.73, residual prediction deviation (RPD) 1.9, root mean square error of prediction (RMSEP) 0.79) was evaluated when a pre-processing step (first derivative) was applied. With a first derivative, R^2 = 0.73, RPD = 2.0 and RMSEP = 0.79 was developed for the equation of chapter 1. In chapter 2 also little differences were found when a first derivative was applied (R^2 = 0.71, RPD = 1.9, RMSEP = 0.90, no pre-processing: R^2 = 0.76, RPD = 2.1, RMSEP = 0.81). These results showed that data pre-treatment (first derivative) could be applied on the equations of chapter 1 and 2, which redues the sensitivity of these equations to baseline shifts.

4.2 Measured NIRS spectra intensity

Pork with low WHC has more protein denaturation (lighter meat, higher colour L* value, increased drip loss%, lower pH) than pork with a high WHC. Denaturated proteins are expected to decrease light penetration, increase scattering and dispersion of light, increase lightness of the meat, and contribute to the paleness of the meat (Beriain et al., 2009). It can be hypothesized that meat with lower WHC might therefore have increased reflection or scatter in comparison to meat with high WHC. In chapter 2 it was evaluated if the NIRS spectra intensity was different between samples with different WHC. Data of chapter two were divided into a low (≥ 6.7% drip loss%), high (≤ 0.6% drip loss%), and moderate (4.0% through 4.1% drip loss%) WHC groups. The average NIRS spectral data of 12 samples per group are shown in figure 1. The absorption value at wavenumber 4875 (2051 nanometres) was selected (NH absorption bands) and revealed that high WHC spectra were significantly different (P = 0.01) from the low WHC spectra. The moderate WHC spectra tended to be different from the high WHC and was not significantly different from the low WHC grouped spectra (P = 0.08 and P = 1.00). NIRS spectra of high WHC (≤ 0.6% drip loss%) samples had higher absorption and thus less reflection compared to samples with low WHC (≥ 6.7% drip loss%). These results are in agreement with measured colour values in chapter 1 to 5, and literature, where meat with lower WHC generates more reflection (Beriain et al., 2009; Mancini & Hunt,

2005). The wave number intensity (Log 1/R) of wave number 4875 correlated significantly with the WHC values of these samples (r = -0.48, *P* = 0.003). Nevertheless, prediction equations developed by partial least squares (PLS) regressions from chapter 2 revealed R^2 values of 0.58 through 0.76 on test-sets and correlated stronger than wave number intensity correlation, and was therefore more preferred for prediction of WHC.

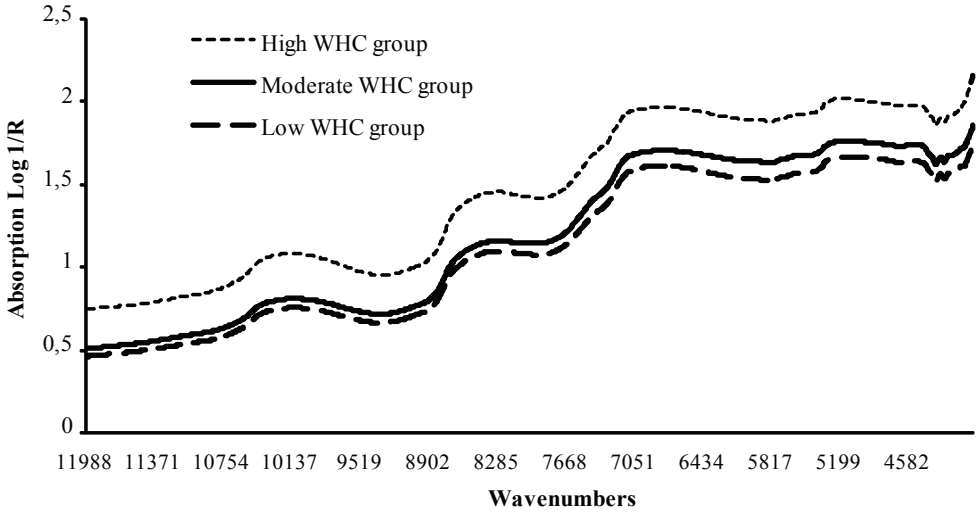

Figure 1. Averaged NIRS spectra of samples (n = 12) grouped by low WHC (≥ 6.7% drip loss%), moderate WHC (4.0 through 4.1% drip loss%), and high WHC (≤ 0.6 % drip loss%).

4.3 Sensitivity

The RPD value indicates the ratio between the standard deviation of reference values and the error of prediction. Root mean square error estimation (RMSEE) is the error for the calibration-set and root mean square error of prediction (RMSEP) is the error of the test-set. The NIRS prediction equations for WHC in chapter 1 revealed a higher RMSEP (0.8) than RMSEE (0.6). Chapter 2 showed that the RMSEP (0.7 through 1.1) values were ≤ RMSEE (0.8 through 1.1) with maximum difference of 0.1. The differences between the RMSEP and RMSEE in chapter 1 and 2 indicated that prediction equations were similar on test-set samples as on the calibration samples. This indicates that the developed prediction equations will perform in a similar way on new independent samples. The RPD values for drip loss% in chapter 1 and 2 ranged from 1.5 through 2.1 on the developed prediction equations, which indicated that the error on the test-set was high in comparison to variations of these test-sets. We assume that reduction of prediction errors might improve the accuracy of WHC prediction by NIRS. Results in chapter 1 revealed that some samples were predicted incorrectly and were therefore sorted in the incorrect WHC groups (22% through 25% was predicted incorrectly), due to the prediction error of the equations. Eventhough, no high drip loss% samples were predicted as having low drip loss%. These results indicated that the developed drip loss% prediction equations were correct, but less sensitive to distinguish moderate WHC pork from high or low WHC pork.

4.4 Specificity

Chapter 5 showed that the NIRS prediction equation on loin spectra could be used to predict WHC of ham muscles from the same carcass (r = 0.25, P <0.05) (chapter 5). Ham muscles are physiologically different muscles in comparison to loin muscles, and results from chapter 1, 2, and 5 indicated that prediction equations by NIRS were carcass specific and could be used on the loin and ham muscles. Adding spectra data from different muscle types into one combined NIRS prediction equation model might further improve specificity for drip loss% prediction by using PLS regressions to eliminate spectra region components that are muscle type dependent. Although NIRS prediction equations are specific for WHC, further research should be performed to investigate data treatment to further improve the NIRS specificity.

4.5 Reproducibility

The sampling procedure of chapter 2 was evaluated for the reproducibility of NIRS measurements and sampling. A total of 4 loins were taken, whereby five slices were sampled from each loin and measured by NIRS. Sampling routine was performed at the pig processing plant (chapter 2). The CV% value for all wave numbers (4000 through 12000 cm^{-1}) of the samples for every loin were measured and averaged. Data revealed that the CV varied between 2% and 16% on reproduced sampling and measurements on pork loins. Pork is a heterogeneous product and on these types of samples CV values ≤16% were considered as acceptable, and can be used for reliable measurements (Petersen et al., 2005). Future research should focus on further minimizing variations during NIRS measurements protocols on pork.

4.6 Repeatability

The repeatability of the NIRS device was tested on the sampling protocol of chapter 2. Multiple NIRS measurements (10x) on 4 different pork loins showed that the NIRS device reproduced similar spectra with very little variation between repeated measurements (CV = 0.20% through 0.03%) on all spectra regions (from 4000 to 12000 cm^{-1}). These results indicated that the NIRS measurement was reliable and had high levels of repeatability on heterogeneous pork.

5. Relevance of NIRS for the pork production chain

Depending on the processing of pork, the required WHC differs to achieve an optimized production process (Bertram et al., 2003; Bryhni et al., 2003; Cariou et al., 1988; Flores et al., 1999; Puolanne et al., 2001; Wismer-Pedersen, 1960). Measuring and sorting fresh meat on WHC can be of interest to optimize the pork supply chain (chapter 5). Cooking of meat induces myofibrils to shrink and proteins to denaturate, thereby enhancing drip loss%, which runs parallel with the pork WHC. Results in chapter 5 showed that WHC values predicted by NIRS significantly correlated with different cooked ham processing yields (tumbling, cooking and final yield) (r ranging from 0.27 through 0.42, $P < 0.001$). When sorting by NIRS, WHC prediction revealed significant difference ($P < 0.05$) between the high and low WHC grouped hams for cooking yield and final yield in chapter 5. The predicted moderate WHC hams were not significantly different from the predicted high WHC hams. These results of chapter 5 were in agreement with literature (Bertram et al., 2003; Bredahl et al., 1998; Bryhni et al., 2003). NIRS prediction equations for WHC can help to optimize cooked ham production.

In addition it can be suggested to treat pork, pre-sorted for WHC, differently during processing to further improve processing yields. The final yield was 4% higher for cooked hams with high predicted WHC values on the fresh pork (highest 20% of all predicted hams) compared to hams with low predicted WHC values (lowest 20% of all predicted hams). These results indicate that pork with a low WHC prediction value can be injected with more brine or with brine containing enhanced water binding properties, to compensate for the low WHC of this pork. Furthermore, fresh pork with high WHC prediction can be used to reduce salt or phosphate levels, because this does not require additional water binding from the brine, which is regarded as beneficial for the public health.

Results in chapter 4 showed no significant ($P > 0.05$) yield differences (injection yield, curing yield, and cooking yield) between pre-selected back bacon, grouped by NIRS WHC predictions. Table 2, chapter 4 indicated that drip loss% did not correlate significantly with injection yield ($r = -0.01$, $P = 0.81$), which suggests that injection yields are less dependent of WHC in back bacon production. The predicted drip loss% of the high WHC group was < 1.47% and for the low WHC group > 3.14%. The data in chapter 4 revealed that overall CV% for injection yield was 11% (mean brine injection% = 16.4 ± 1.8) for experiment 2. The prediction errors of NIRS for WHC in chapter 2 revealed that the RMSEP varied from 0.8 through 1.1. This means that the prediction error was almost similar to the SD (1.8) of the injection yield values. The combination of a low CV value of 11% and no significant correlation between drip loss% and injection yield indicate that significant differences between the NIRS pre-selected WHC groups were very unlikely. Nevertheless, the pHu and colour L* was significant different ($P < 0.05$) between the high and low WHC predicted groups in chapter 4 and showed that NIRS predictions were able to differentiate in pork quality. However, NIRS WHC prediction for optimizing back bacon production was regarded as not useful.

The required NIRS scan time is important for rapid prediction of pork WHC. Chapter 4 revealed that changing the resolution from 16 cm^{-1} to 32 cm^{-1} reduced the scan time from the NIRS measurements from ~ 10 sec through ~ 3.8 sec. This resulted, however, in unacceptable prediction equations with R^2 of 0.50 that was within the range of other studies (0.31 – 0.79) (Prieto et al.,

2009). Results in chapter 4 suggest therefore that a scan time of 3.8 seconds could not be used for accurate measurements, or should be improved with further research. A scan time of approximately 3.8 seconds allows a maximum of 947 pork samples to be scanned per hour, while a scan time of 10 seconds allows 360 pork samples to be scanned per hour. The prediction equation, developed with a 10 second scan time was found to be suitable for a practical application (chapter 2 and 5) and can still be regarded as rapid prediction of WHC.

6. General conclusions

Currently used parameters to predict pork WHC (pH, colour L*, and drip loss%) were regarded as low correlating, invasive, time consuming, and not applicable for rapid prediction of pork WHC. This thesis showed that NIRS prediction equations for WHC could be developed for pork loin samples measured at pig processing plants. In addition, early post mortem moisture absorption could be used for prediction and indication of drip loss% at 24 hours but this was not regarded as rapid and applicable under commercial conditions at a pig processing plants.

Predictions of pork WHC by NIRS could be used to optimize processing yields of cooked ham production. However, NIRS prediction of WHC could not be used to optimize back bacon production yields. Measurements of NIRS are applicable for rapid prediction of pork WHC, even though prediction equations could be further improved.

Further research could be performed to study the relation of pork quality to processing conditions of fresh and further processed meat products. Application of rapid online WHC measurements might be linked to existing carcass information, whereby implications for the primary production, such as genetics, husbandry, animal handling, and feeding strategies can be investigated. The pork supply chain can further optimize fresh and processed pork by sorting for WHC. Research on logistical and decision support models should be carried out to allow practical application of sorting on pork WHC to optimize the pork supply chain.

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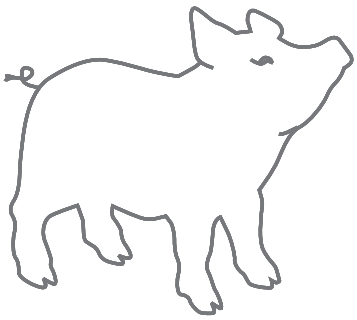


Summary

The aim of this thesis was to validate rapid prediction of pork water holding capacity. Correlations between fresh pork measurements and pork water holding capacity and its technological quality were investigated. In addition application of NIRS technology to predict pork WHC, and its correlation with further processing yields of some pork products was investigated. Water holding capacity (WHC) of pork defines the sensory appreciation and processing yields of meat (chapter 1, 2, and 5). Pork is a heterogeneous product which has a natural variation in water holding capacity (chapter 1 to 5). This variation in WHC is generated by differences in post mortem muscle metabolism of carcasses. Differences in post mortem metabolism depend on peri mortem factors, such as the amount of available muscle energy storages, genetics, and stress of the animal during slaughter. The speed of post mortem muscle metabolism affects the amount of generated lactic acid and thereby affects protein denaturation, aggregation, enzymatic protein regeneration, and rigor in the muscle. Changes in these muscle proteins largely affect the protein water binding properties by hydrogen binding, spatial, and capillary binding post mortem (chapter 3). Variation in pork WHC still occurs within current pig processing plants eventhough extensive research has identified numerous factors that affect pork WHC. Nowadays, the pork processing industry has been specialized in sorting of carcasses and primal cuts on the basis of weight and lean characteristics. Additional sorting of pork by WHC can further optimize processing yields (chapter 1, 2, and 5). The rapid prediction and sorting for WHC is currently limited at pork processing plants.

The first objective of this thesis was to investigate the possibilities of a rapid prediction of pork WHC by measuring parameters such as pH, colour L*, drip loss%, water absorption, and by NIRS at laboratory scale and at pig processing plant scale (chapter 1, 2, and 3). The second objective was to investigate if predictions of pork WHC could be used to optimize processing of pork (chapter 4 and 5). It was first investigated if NIRS could predict pork quality of loin slices (pH, colour L*, and drip loss%), and how the NIRS parameters correlated with the different pork quality measurements. Drip loss% after 48 hours storage in a retail case-tray was used to determine pork WHC, whereby low drip loss% values represent high pork WHC.

In chapter 1, prediction of loin pork quality by NIRS measurements was investigated at laboratory conditions. Prediction equations were developed for drip loss% (WHC), colour L*, a*, b*, and pH ultimate (pHu). For drip loss% value a prediction equation was developed (R^2 0.73, RPD 1.9). Pork samples grouped on the WHC value predicted by NIRS, were grouped as superior (drip loss < 2%) and inferior (drip loss > 4%), and revealed that 76% of the samples were predicted in the correct group. For colour L*, test-set samples were predicted with R^2 0.75, RPD 2.0, colour a* R^2 0.51, RPD 1.4, colour b* R^2 0.55, RPD 1.5, and pHu R^2 0.36, RPD 1.3. Results of chapter 1 revealed that NIRS prediction equations could be developed to predict drip loss% and colour L* of pork samples. However, NIRS equations for colour a*, b*, and pHu were not applicable for prediction of pork quality of commercially processed pigs.

The positive results of NIRS to predict WHC and colour L* at laboratory scale led to further research to study NIRS prediction of pork quality (pH, colour L*, and drip loss%) under pig processing plant conditions (chapter 2). Pork loin slices from four different pig processing plants

were used to develop prediction equations for pork quality with NIRS. Equations with $R^2 > 0.70$ and residual prediction deviation (RPD) ≥ 2.0 were considered as applicable for screening. Production plant A showed R^2 0.76 and RPD 2.05, other plants (B, C, and D) showed $R^2 < 0.70$ and RPD < 2.0 for drip loss%, which indicated that prediction equations were not adequate for predictions of drip loss%. Samples were grouped by predicted drip loss% values; superior (< 2.0%), moderate (2-4%), inferior (> 4.0%). The results from grouping by NIRS predicted drip loss% revealed that 64% from the superior group and 56% from inferior group were predicted correctly. Only prediction equation from plant A could be used for screening and prediction of drip loss%. The prediction equations for colour L*, and pHu did not meet the requirements (R^2 0.70 and RPD 1.82), and were regarded as not useful. In chapter 2 it was concluded that NIRS prediction equations can be used for screening drip loss% of pork, which is measured at pig processing plants.

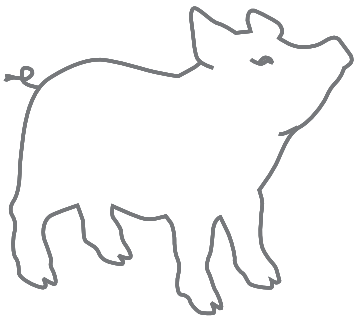
Water holding capacity is the ability of meat to hold moisture and is subjected to events occurring in muscle tissues early post mortem. Post mortem lactic acid accumulation and thereby provoked protein denaturation, aggregation, and rigor are seen as processes which affect pork WHC. The objective of chapter 3 was to characterize the loss of moisture from muscle post mortem and investigate whether moisture losses are useful in predicting drip loss% of fresh pork. Cotton-rayon absorptive material were inserted in the *Longissimus dorsi* muscles of pork carcasses (n = 51) immediately (from 15 minutes after exsanguination) post mortem and removed at 15 minutes intervals or remained in the muscle for 24 hours. Drip loss% varied (0.6 to 15.3%) across carcasses. The highest moisture absorption was observed at 105 minutes post exsanguination after 15 minutes dwell time. Individual absorption at 75 minutes post mortem (15 minutes dwell time) correlated with final drip loss% (r = 0.33). Correlations were highest by individual absorption values at 90 minutes post mortem (r = 0.48) and accumulated absorption values at 150 minutes post mortem (r = 0.41) during the first five hours post mortem. Results of chapter 3 reveal moisture loss from muscle tissue early post mortem and suggest that select time periods post mortem correspond to culmination of biochemical and physical events facilitating moisture release, which can be used for early drip prediction. In addition, results of chapter 3 suggest an approach for capturing moisture release early post mortem is possible and may be used to predict WHC in fresh pork. However, early post mortem moisture absorption is regarded as less applicable for rapid non-invasive WHC prediction.

Based on the results from chapter 1, 2, and 3, NIRS prediction equations were seen as best applicable for rapid prediction of pork WHC. Therefore it was investigated if fresh pork quality parameters and NIRS measurements on fresh back bacon primals correlated to technological yields during back bacon production. Chapter 4 investigated if NIRS could be used to optimize back bacon production and if NIRS prediction equations could be developed for prediction technological yields of back bacon. Two experiments were conducted on pork middles from 356 (experiment 1) and 329 (experiment 2) carcasses and fresh meat quality (pHu, colour L*, drip loss%, and NIRS predicted WHC) and processing yields were measured. Ultimate pH and colour L* had low, but significant correlations ($r \leq 0.26$) with technological back bacon yields. Developed NIRS prediction equations for drip loss% resulted in low R^2 values (0.50), while technological yields could not be predicted ($R^2 < 0.21$ and RPD < 1.1). Preselection of back bacons by NIRS predicted WHC values in experiment 2, did not result in significant differences on fresh meat quality parameters, injection yield, and yield

before frozen storage between the groups. Differences in production yields between both NIRS predicted WHC groups were not significant, however pHu and colour L* was significant different between both NIRS pre-selected back bacon groups. The results in chapter 4 revealed that back bacon length and storage of back bacons during curing significantly affected processing yields. Correlation between fresh meat quality parameters (pHu, colour L*, and drip loss%) and injection yield was not significant.

During pork cooking processes, proteins are affected by heat treatment, which provokes protein denaturation and aggregation, and thereby affects WHC of cooked products. Therefore in chapter 5 it was investigated if parameters, such as pH, colour L*, drip loss%, and NIRS WHC predicted values of fresh pork primals correlated to yields during cooked ham production. NIRS measurements on fresh pork ham primals were investigated if they could be used to predict cooked ham production yields. Fresh meat quality (pHu, colour L* value, drip loss%) were measured on loins and hams from 160 pigs, randomly selected at one production plant. Cooked ham yields (transport loss, injection, and tumbling yield) were determined on individual or combined silverside and topside muscles (cooking and final yield). Ultimate pH from ham correlated significantly with all yield values ($r = -0.19$ to $r = -0.55$). NIRS prediction on ham had highest correlation for cooking ($r = -0.42$) and final yield ($r = 0.37$) in comparison to colour L*. Cooking yield correlated significantly with colour L* ($r = -0.20$) and pHu value of the loin ($r = 0.23$). Transport loss of ham muscles correlated significantly with cooking ($r = -0.36$) and final yield ($r = 0.35$), while drip loss% of loin samples also correlated significantly with cooking ($r = -0.32$) and final yield ($r = 0.36$). Pre-selection of hams by NIRS into three predicted WHC (drip loss%) groups resulted in differences between the low, moderate, and high WHC groups for cooking and final yield. It was concluded that NIRS can be used to predict rapid fresh ham quality for sorting and optimization of the cooked ham process.

The overall conclusion of this thesis is that currently used parameters to predict pork WHC (pH, colour L*, and drip loss%) are regarded as not applicable for rapid prediction of pork WHC. Early post mortem moisture absorption can be used for prediction and indication of drip loss% at 24 hours but this is not regarded as rapid and applicable at a pig processing plants. This thesis concludes that NIRS prediction equations for WHC can be developed for pork loin samples measured at pig processing plants and can be used to optimize processing of pork.



Samenvatting

Het doel van dit proefschrift was om een snelle voorspelling van het waterhoudend vermogen van varkensvlees te valideren. Correlaties tussen metingen aan varkensvlees, waterhoudend vermogen en de betreffende verwerkingsrendementen werden onderzocht. Bovendien werd de toepasbaarheid van NIRS (nabij infrarood spectroscopie) op varkensvlees om het waterhoudend vermogen (WHC) te voorspellen onderzocht. Ook werden correlaties tussen waterhoudend vermogen en de verwerkingsrendementen van bacon en hammen onderzocht. Waterhoudend vermogen van varkensvlees bepaalt in grote mate de sensorische waardering en de verwerkingsrendementen van vleesproducten (hoofdstuk 1, 2, en 5). Varkensvlees is een heterogeen product welke een natuurlijke variatie in waterhoudend vermogen heeft, waardoor tussen karkassen variatie in waterhoudend vermogen kan worden aangetroffen (hoofdstuk 1 tot en met 5). Deze variatie in waterhoudend vermogen wordt gegenereerd door verschillen in post mortem spiermetabolisme tussen karkassen. Verschillen in post mortem metabolisme zijn afhankelijk van perimortem factoren, zoals de hoeveelheid beschikbare energievoorraad in de spieren, genetica en de stress die het varken ondervindt tijdens het slachten. De snelheid van post mortem spiermetabolisme beïnvloedt de hoeveelheid gegenereerd melkzuur waardoor de snelheid en mate van eiwit denaturatie, aggregatie, regeneratie door enzymatische eiwitten en de uiteindelijke rigor mortis in de spier wordt bepaald. Veranderingen van deze spiereiwitten na het slachten beïnvloeden in grote mate de eiwit- en waterinteractie (binding) door bijvoorbeeld de aanwezigheid van waterstofionen en de ruimtelijke en capillaire bindingen (hoofdstuk 3).

Ondanks dat door uitgebreid onderzoek de factoren die waterhoudend vermogen bepalen zijn geïdentificeerd, komt bij de huidige varkensslachterijen nog steeds variatie in waterhoudend vermogen tussen karkassen voor. Tegenwoordig hebben varkensslachterijen zich gespecialiseerd in het sorteren van karkassen en deelstukken op basis van gewicht en vet kenmerken. Echter, een snelle voorspelling van waterhoudend vermogen en sorteren van karkassen op basis van waterhoudend vermogen is momenteel niet mogelijk voor varkensslachterijen. Het aanvullend sorteren van varkensvlees op basis van waterhoudend vermogen kan de verwerking en technologische opbrengsten optimaliseren (hoofdstuk 1, 2, en 5).

Een snelle voorspelling van waterhoudend vermogen op varkensvlees is gemeten, waarbij pH, kleur L*, dripverlies%, waterabsorptie, en NIRS zijn onderzocht. De metingen werden uitgevoerd onder laboratoriumcondities en onder varkensslachterijcondities (hoofdstuk 1, 2 en 3). De tweede doelstelling was om te onderzoeken of voorspellingen van waterhoudend vermogen van varkensvlees gebruikt kunnen worden om de verwerking van varkensvlees verder te optimaliseren (hoofdstuk 4 en 5). Allereerst werd onderzocht of NIRS de kwaliteit van de rugspier (*Longissimus dorsi*) (pH, kleur L*, en dripverlies%) kan voorspellen en hoe de NIRS parameters correleren met de kwaliteitsparameters van varkensvlees. Dripverlies% na 48 uur opslag in een retailverpakking werd gebruikt om waterhoudend vermogen van varkensvlees te bepalen, waarbij lage dripverlies% een hoog waterhoudend vermogen vertegenwoordigt.

In hoofdstuk 1 werd een kwaliteitsvoorspelling van de *Longissimus dorsi* met behulp van NIRS metingen onderzocht onder laboratorium omstandigheden. Voorspelmodellen werden

ontwikkeld voor dripverlies% (waterhoudend vermogen), kleur L*, a*, b* en uiteindelijke pH (pHu). Voor dripverlies% werd een voorspellende vergelijking ontwikkeld (R^2 0,73, residual prediction deviation (RPD) 1,9). Varkensvlees monsters, gegroepeerd op basis van het door NIRS voorspelde waterhoudend vermogen, werden onderverdeeld in 2 dripverlies klassen; superieur (dripverlies <2%) en inferieur (dripverlies> 4%). Hierbij werd aangetoond dat 76% van de monsters in de juiste groep waren voorspeld. Voor kleur L*, werden test-set monsters voorspeld met R^2 0,75, RPD 2.0, kleur a* R^2 0,51, RPD 1.4, kleur b* R^2 0,55, RPD 1,5, en pHu R^2 0,36, RPD 1.3. Resultaten van hoofdstuk 1 tonen aan dat NIRS kan worden toegepast om waterhoudend vermogen en kleur L * van varkensvlees monsters te voorspellen. Echter, de ontwikkelde NIRS vergelijkingen voor kleur a *, b *, en pHu waren niet geschikt voor de voorspelling van de kwaliteit van commercieel verwerkt varkensvlees.

De positieve resultaten van NIRS voor de voorspelling van waterhoudend vermogen en kleur L* onder laboratorium condities heeft geleid tot verder onderzoek naar de voorspelling van varkensvlees kwaliteit (pH, kleur L *, en dripverlies%) met behulp van NIRS onder de praktijk-omstandigheden bij varkens slachterijen (hoofdstuk 2). *Longissimus dorsi* monsters van karkassen gemeten in vier verschillende varkensslachterijen, werden gebruikt om voorspelmodellen voor NIRS te ontwikkelen. Vergelijkingen met $R^2 > 0,70$ en RPD $\geq 2,0$ werden beschouwd als toepasbaar voor het doorlichten van vlees met behulp van NIRS onder praktijkomstandigheden. Voorspelmodellen voor NIRS met data van locatie A toonden aan dat de vergelijking toepasbaar was (R^2 0,76 en 2,05 RPD), echter resultaten van andere productielocaties (B, C, en D) toonden $R^2 <0,70$ en RPD <2.0 voor dripverlies%, welke niet geschikt waren voor het voorspellen van dripverlies% onder praktijkomstandigheden. Hierdoor kon alleen voorspellingsvergelijking van locatie A worden gebruikt voor het voorspellen van dripverlies%. Varkensvlees monsters van locatie A, gegroepeerd op basis van de met behulp van NIRS voorspelde waarde voor waterhoudend vermogen, werden onderverdeeld in 3 dripverlies klassen; superieur (<2,0%), matig (2-4%), inferieur (> 4,0%). De resultaten toonden aan dat 64% van de superieure groep en 56% van inferieure groep correct werden voorspeld. De voorspelmodellen voor kleur L*, en pHu voldeden niet aan de eisen (R^2 0,70 en RPD 1,82) en werden beschouwd als niet zinvol. In hoofdstuk 2 is geconcludeerd dat NIRS voorspelmodellen gebruikt kunnen worden voor het voorspellen van dripverlies% gemeten onder praktijkomstandigheden.

Waterhoudend vermogen is het vermogen van vlees om vocht vast te houden en is, zoals eerder aangegeven in deze samenvatting, onderworpen aan gebeurtenissen die zich afspelen in spierweefsel vroeg post mortem. De accumulatie van post mortem melkzuur en de daardoor uitgelokte eiwit denaturatie, aggregatie, en rigor mortis van de spier worden gezien als processen die het waterhoudend vermogen van varkensvlees beïnvloeden. Het doel van hoofdstuk 3 was om het post mortem vochtverlies uit de spier te karakteriseren en te onderzoeken of de hoeveelheden vochtverlies nuttig zijn bij het voorspellen van dripverlies% van vers varkensvlees. Absorberend materiaal (katoen) werd onmiddellijk post mortem (vanaf 15 minuten na afbloeden) in *Longissimus dorsi* spieren van varkens karkassen (n = 51) ingebracht en verwijderd met een tussentijd van 15 minuten gedurende 300 minuten post mortem of bleven in de spier gedurende 24 uur. Dripverlies% varieerde tussen karkassen (0,6 tot 15,3%). De meeste vochtopname door het absorberend materiaal

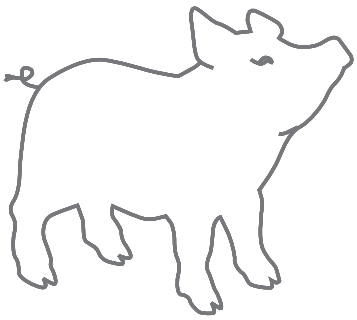
werd waargenomen op 105 minuten na verbloeding. Individuele absorptie op 75 minuten post mortem werd gecorreleerd met uiteindelijke dripverlies% (r = 0,33). Correlaties waren het hoogst bij individuele absorptiewaarden na 90 minuten post mortem (r = 0,48) en geaccumuleerde absorptiewaarden op 150 minuten (r = 0,41) gedurende de eerste vijf uur post mortem. Resultaten van hoofdstuk 3 suggereren dat vochtverlies uit spierweefsel vroeg post mortem (< 5 uur na afbloeden), overeenkomt met biochemische en fysische gebeurtenissen welke vochtuittreding beïnvloeden, en daardoor gebruikt kunnen worden voor vroege voorspelling van dripverlies. Daarnaast suggereren resultaten van hoofdstuk 3 dat het vastleggen van het vrijgekomen vocht met behulp van absorberend materiaal vanuit het spierweefsel vroeg post mortem mogelijk is en kan worden gebruikt om waterhoudend vermogen te voorspellen op vers varkensvlees. Echter, de vroege post mortem vochtopname werd beschouwd als een methode welke minder geschikt is voor snelle praktische (niet-invasieve) methode voor de voorspelling van het waterhoudend vermogen.

Gebaseerd op de resultaten uit hoofdstuk 1, 2 en 3 werden NIRS voorspelmodellen gezien als best toepasbaar voor een snelle voorspelling van het waterhoudend vermogen van varkensvlees. Daarom werd onderzocht of vers varkensvlees kwaliteitsparameters en NIRS metingen aan verse back bacon deelstukken (*Longissimus dorsi*) correleerden met de verwerkingsrendementen tijdens back bacon productie. In hoofdstuk 4 werd onderzocht of NIRS gebruikt kon worden om bacon productie te optimaliseren en of NIRS voorspelmodellen ontwikkeld konden worden voor de voorspelling van de verwerkingsrendementen tijdens de productie van bacon. Twee experimenten werden uitgevoerd aan deelstukken van varkens (middels). In totaal werden aan 356 (experiment 1) en 329 (experiment 2) karkassen vers vlees kwaliteit (pHu, kleur L *, dripverlies% en NIRS voorspelde waterhoudend vermogen) en verwerkingsrendementen gemeten. Uiteindelijke pH en kleur L * hadden lage, maar significante correlaties ($r \leq 0,26$) met de verwerkingsrendementen (experiment 1). De ontwikkelde NIRS voorspelmodellen voor dripverlies% resulteerden in lage R^2 waarden (0,50), terwijl de verwerkingsrendementen niet konden worden voorspeld (R^2 <0,21 en RPD <1.1). Voorselectie van back bacons op waterhoudend vermogen met behulp van de NIRS voorspelde waarden (experiment 2) heeft niet geleid tot significante verschillen van enkele verwerkingsrendementen (injectie opbrengst en opbrengst voor bevroren opslag) tussen de groepen, maar pHu en kleur L * waren wel significant verschillend tussen beide NIRS voorgeselecteerde bacon groepen. Uit de resultaten in hoofdstuk 4 is gebleken dat baconlengte en de opslag van back bacons tijdens het egaliseren (gedurende enkele dagen de geïnjecteerde pekel in de back bacons laten diffunderen) de verwerkingsrendementen sterk beïnvloeden. Er zijn geen significante correlaties gevonden tussen vers vlees kwaliteitsparameters (pHu, kleur L*, en dripverlies%) en injectie opbrengst.

Tijdens de bereidingsprocessen van varkensvlees worden de eiwitten beïnvloed door de warmtebehandeling, welke eiwit denaturatie en aggregatie veroorzaken, die vervolgens het waterhoudend vermogen van gekookte producten beïnvloeden. Derhalve werd in hoofdstuk 5 onderzocht of parameters zoals pH, kleur L*, dripverlies% en waterhoudend vermogen van verse varkenshammen voorspeld met NIRS, correleerden met de verwerkingsrendementen tijdens de gekookte ham productie. Er werd onderzocht of NIRS metingen aan verse varkenshammen konden worden toegepast om de verwerkingsrendementen van gekookte ham te voorspellen. Vers vlees kwaliteit (pHu, kleur L*, en dripverlies%) werden gemeten op *Longissimus dorsi* en

de hammen van 160 varkens, welke willekeurig werden geselecteerd op een productielocatie. De verwerkingsrendementen van de gekookte hammen (transport verlies, injectie, en tumbling opbrengsten) werden bepaald op individuele of gezamenlijke plattebil (*Biceps femoris*) en bovenbil (*Semimembranosus*) spieren (kookopbrengst en uiteindelijke opbrengst). De pHu van hammen correleerde significant met alle opbrengstwaarden (r = -0,19 tot r = -0,55). NIRS voorspelling (dripverlies) op ham had hoogste correlatie met kookopbrengst (r = -0,42) en uiteindelijke opbrengst (r = 0,37) in vergelijking met kleur L*. Kookopbrengst correleerde significant met kleur L* (r = -0,20) en pHu van de *Longissimus dorsi* (r = 0,23). Transportverlies van de hamspieren correleerde significant met de kook opbrengst (r = -0,36) en de uiteindelijke opbrengst (r = 0,35), terwijl dripverlies% van de *Longissimus dorsi*-monsters ook significant correleerde met de kookopbrengst (r = -0,32) en de uiteindelijke opbrengst (r = 0,36). Pre-selectie van hammen met NIRS in drie voorspelde waterhoudend vermogen (dripverlies%) groepen resulteerde in verschillen tussen de lage, matige en hoge waterhoudend vermogen groepen voor de kookopbrengst en de uiteindelijke opbrengst. Uit dit experiment werd geconcludeerd dat de toepassing van NIRS gebruikt kan worden om snel de verse hamkwaliteit te voorspellen, te sorteren en om de productie van kookhammen te optimaliseren.

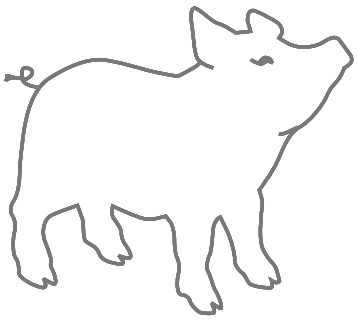
De algemene conclusie van dit proefschrift is dat de op dit moment gebruikte parameters om waterhoudend vermogen van varkensvlees (pH, kleur L*, en dripverlies%) te voorspellen beschouwd kunnen worden als niet toepasbaar voor een snelle voorspelling van waterhoudend vermogen van varkensvlees. De vroege post mortem vochtopname met behulp van absorberend materiaal aan karkassen kan gebruikt worden voor de voorspelling en indicatie van dripverlies% na 24 uur, maar dit wordt echter niet beschouwd als snel en toepasbaar voor slachterijen. In dit proefschrift is aangetoond dat NIRS voorspelmodellen ontwikkeld kunnen worden op de *Longissimus dorsi* voor het voorspellen van het waterhoudend vermogen. Deze modellen kunnen gebruikt worden om de varkensvleesverwerking te optimaliseren.



Curriculum Vitae

Christiaan Kapper werd geboren op 29 maart 1980 in Warnsveld. Hij is daar opgegroeid tot zijn tiende levensjaar. In 1990 is het gezin naar Westeremden, Groningen verhuist. In 1998 behaalde Christiaan zijn HAVO diploma op het Ommelander College te Appingedam, en startte met zijn studie aan de CAH te Dronten. Tijdens zijn studie veehouderij in Dronten heeft hij gekozen voor de specialisatie veevoeding. Gedurende zijn studie in Dronten heeft Christiaan 6 maanden stage gelopen in Zuid Afrika, waar hij mogelijkheden voor de verbetering van de uiergezondheid van melkkoeien in Zuid Afrika heeft onderzocht. Hij heeft stage gelopen in Kwasizabantu, Kwazulu Natal en Tsitsikamma, Port Elisabeth. In 2002 begon Christiaan aan de vervolgstudie Dierwetenschappen aan Wageningen Universiteit in Wageningen. Hij koos voor de studierichting Dierlijke Productie Systemen en is afgestudeerd met specialisatie Adaptatie Fysiologie. Voor Adaptatie Fysiologie deed hij onderzoek naar de fysiologische functie van melkvet variatie tijdens het melken van koeien.

Na het afstuderen in 2004 heeft Christiaan twee jaar gewerkt voor Wageningen Centre for Food Science (WCFS) als onderzoeksassistent bij de groep Integration of texture and flavour perception. In 2006 besloot Christiaan om gedurende één jaar te werken aan een pilot voor het gebruik van elektronische oormerken en elektronische dierregistratie voor schapen en geiten bij het ministerie van Economische zaken, Landbouw en Innovatie (EL&I) Vanaf september 2007 is Christiaan begonnen als promovendus aan dit proefschrift voor VION Food Group, welke in samenwerking met Nutricontrol was opgezet. Vanaf september 2011 is Christiaan begonnen als onderzoeker bij VION Fresh Meat North GmbH in Düsseldorf, waar hij werkt binnen een Marie Curie project: Quality and Risk Management in Meat Chains (QUARISMA).



Training and Supervision plan

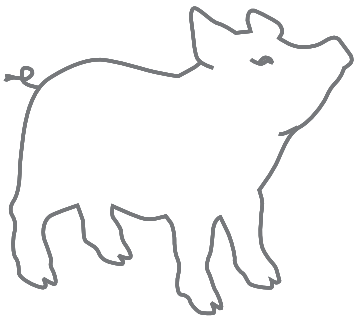
Graduate School WIAS

Name	Christiaan Kapper
Group	Animal nutrition
Project term	2007 - 2012
Supervisors	Dr. ir. R.E. Klont (Daily), Dr. Ir. J.M.A.J. Verdonk, Prof. Dr. Ir. H.A.P. Urlings

The Basic Package	**year**	**credits ***
WIAS Introduction Course	2008	1,5
WIAS Course on philosophy of science and ethics	2008	1,5
International conferences		
QPorkChains, Barcelona, Spain	2007	0,9
ICoMST 2008, Cape Town, South Africa	2008	1,5
IDRC 2010 “chambersburg conference”, Chambersburg, USA	2009	1,5
NIR2011, Cape Town, South Africa	2011	1,5
QPorkChains, Mallorca, Spain	2011	1
Seminars and workshops		
Workshop “An introduction in the application of NIRS”	2007	0,6
Seminar “Spierontwikkeling en vleeskwaliteit”	2007	0,3
WIAS Science day (2x)	2008-9	0,6
QporkChains workshop	2008	0,3
PhD workshop NVG	2010	0,3
Presentations		
ICoMST, Cape Town, poster	2008	1,0
Q PorkChains, Wageningen, oral	2009	1,0
Q PorkChains, Budapest, oral	2009	1,0
WIAS Science day, Wageningen, poster	2009	1,0
Q PorkChains, Bonn, poster	2009	1,0
NIR 2011, Cape Town, oral	2011	1,0
Q PorkChains, Mallorca, oral	2011	1,0
In-depth Studies		
Advanced meat course	2009	4,0
Optimising NIR method; get the sampling and spectroscopy right	2011	0,3
Orientation on mathematical modelling in biology	2011	1,5
Design animal experiments	2009	1,0

Professional Skills Support Courses		
Project- and Time Management	2008	1,5
Techniques for writing and presenting scientific papers	2008	1,2
Scientific writing	2011	1,8
Preparing PhD research proposal	2007	6,0
External training period, 6 months, Virginia Tech University, USA	2010	2,0
Introduction to R	2009	0,6
FT-IR training course	2009	0,6
Didactic skills training		
Supervisor BSc Course: Introduction to Animal Science	2011	0,5
Supervisor BSc. Theses and Internships (8 students)	2009-12	4,0
Management skills training		
Young Employees Cehave (YEC)	2009	2,0
Education Committee	2010-2011	2,0
Total		**48**

** one ECTS (European Credit Transfer System) credit equals a study load of approximately 28 hours.*



Colophon

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