EVALUATION OF PORK MEAT QUALITY BY USING WATER HOLDING CAPACITY AND VIS-SPECTROSCOPY

By

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ABSTRACT

The goal of this research was to investigate the use of water holding capacity (WHC) and vis-spectroscopy to classify pork meat quality. This study was carried out in two stages.

In the first part, the suitability of using different WHC measuring methods (bag method at 2 and 4 days, centrifuge, cotton-rayon material and filter paper methods) to classify the pork meat samples were studied. The methods were compared to see which method was able to discriminate pork meat samples according to their defined quality classes. The meat samples were grouped into 4 quality classes, namely PFN (pale, firm and non-exudative), PSE (pale, soft and exudative), RFN (red, firm, and non-exudative), and RSE (red, soft, and exudative). The discriminant analysis using stepdisc was used to separate the quality groups. Cotton-rayon material and filter paper methods were better than the other WHC measuring methods to classify FN (Firm, Non-exudative) and SE (Soft, Exudative) groups.

In the second stage, the aim was to investigate visible spectroscopy for the classification of different pork meat quality classes. Discriminant procedure was performed for grouping quality classes and stepdisc was used to select the suitable wavelengths. The results showed that it was possible to separate the P (Pale) classes of pork meat samples from the R (Red) classes of pork meat samples with an accuracy of about 85 % and chosen wavelengths were 500, 430, 550, 570 and 510 nm.

RÉSUMÉ

Cette étude a visé l'évaluation de la capacité de rétention d'eau (CRE) et la spectroscopie en spectre visible, pour l'évaluation de la qualité de la viande porcine.

En premièr lieu, différentes méthodes pour mesurer la CRE (suspension et égouttement pour 2 ou 4 jours, centrifugation, absorption par matériau coton-rayone, ou par papier filtre), servant à classifier les échantillons de viande porcine selon des critères de qualité bien définis, furent comparées. Les échantillons de viande porcine furent regroupés en quatre classes de qualité: PFN (pâle, ferme et non-exudative), PSE (pâle, mol et exudative), et RFN (rouge, ferme et non-exu). Une analyse discriminante utilisant l'option STEPDISK servit à séparer ces quatre classes de qualité. Pour discriminer entre les viandes FN (ferme, non-exsudatif) et SE (mou, exsudatif), les méthodes de mesure de la CRE par absorption avec coton-rayone ou papier filtre furent les plus performantes.

En deuxiéme lieu phase, une classification de la qualité de la viande porcine par spectroscopie en spectre visible fut visée. L'analyse discriminante servit à regrouper les échantillons en catégories de qualité, puis l'option STEPDISK a sélectionnée les longueurs d'ondes les plus appropriées. En choisissant des longueurs d'ondes de 500, 430, 550, 570, et 510 nm, il fut possible de distinguer, avec une exactitude de 85%, entre les classes P (pâle) et R (rouge) de viande porcine.

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CONTRIBUTION OF AUTHORS

The work reported here was completed by the candidate and was supervised by Dr. Michael Ngadi of the Department of Bioresource Engineering, Macdonald Campus of McGill Univeristy, Montreal. The entire experiments were carried out at the Food Engineering Laboratory at McGill University.

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NOMENCLATURE

a^*	The component from green to red
b^*	The component from blue to yellow
CMV	Colour Machine Vision
CPC	The Canada Pork Council
DA	Discriminant Analysis
DFD	Dark purplish red, very Firm and Dry
DHA	Docosahexaenoic
EU	European Union
EPA	Eicosapentaenoic
FN-SE	Firm-Non-exudative-Soft Exudative
GLM	General linear model
IMF	Intramuscular fat
JPCS	The Japanese Pork Color Standards
L^*	Brightness
LD	*M.longissimus dorsi*
MHz	Mega hertz
NIRS	Near Infrared Spectroscopy
nm	Nanometers
NPPC	The National Pork Producers Council
PCA	Partial Component Analysis
PFD	Pale, Firm, Dry
PFE	Pale, Firm, Exudative
PFN	Pale, Firm, Non-exudative
PM	*M.psoas major* or *psoas* muscle from the 'fillet'
PSE	Pale pinkish gray, very Soft and Exudative
PSS	Porcine Stress Syndrome
RFN	Reddish pink, Firm and Non-exudative
RSE	Red, soft and exudative
US	United States

USA	The United States of America
USDA	The United States Drug Adminitration
WHC	Water-holding capacity

CHAPTER I. GENERAL INTRODUCTION

According to Canada Pork International, Canada is the world's third largest exporter of pork meat after the US and EU. Accurately predicting the pork meat quality is very crucial to success in the meat industry. When accurate quality is not achieved, the pork processors and the pork producers suffer economic losses (Chan *et al.,* 2002). For instance, when high quality, highly palatable pork is processed instead of being sold as fresh pork to the consumer, or when lower quality fresh pork is sold and found unsatisfactory by the consumers, both situations result in economic losses and therefore, have an impact on business success.

Competition with domestic and international markets provides higher motivation for improving the quality of pork produced in Canada, the US as well as EU. Accurate prediction of quality, allows meat packers to minimize economic losses during processing (Tan *et al.,* 2000).

Traditionally, pork quality can be classified into four categories based on color, texture (firmness), and exudation (drip loss). They are RFN, PSE, DFD and RSE. The first quality group is referred to as RFN (Reddish pink, Firm and Non-exudative), which is the classification for the best quality pork. RFN meat has desirable colour, firmness, normal water holding capacity (WHC), and moderate decline rate of pH. RFN has a normal ultimate pH (5.8-5.6). The second group is PSE (Pale pinkish grey, very Soft and Exudative) which refers to meat that has an undesirable appearance and shrinks excessively. PSE classes of meat have very poor water-holding capacity (WHC) and a rapid decline rate of pH (5.6-5.5). The third group is DFD (Dark purplish red, very Firm and Dry) meat. DFD meats have firm and sticky surface with high WHC and very high pH. Generally DFD meat occurs as a result of long-term stress from improper handling of live animals (National Pork Board, 1999). DFD meat is frequent in cattle although it can occur in pigs, too. The DFD meat has a high ultimate pH, which represents a hygienic risk and lowers its storability (Hambrecht, 2004). The last one is RSE, which is red, soft and exudative pork meat. One of the major limitations to predict the pork quality has been the presence of RSE pork (Kauffman *et al.*, 1992; Warner, 1994). Since the color is similar to RFN pork, most chemical or physical procedures have failed to

differentiate RSE from RFN. Beside the four group of pork meat, there are also other quality classes defined as PFN (Pale, Firm, Non-exudative), PFE (Pale, Firm, Exudative), and PFD (Pale, Firm, Dry) in literature. In the study of Kauffman et al. (1992), new quality categories were recommended that included RSE and PFN because some samples were pale but had low drip loss, whereas considerable numbers were reddish pink but had high drip losses. Kanda and Kancchika (1992) observed PFE and PFD pork. Also Roseriro et al. (1993) reported that many red or reddish-pink samples had unacceptable drip loss whereas some pale samples would not exudate excessively after 24 hr. Furthermore, they observed that some dark samples had unacceptable drip loss. However, these quality classes are of less importance because of their low frequency (Hambrecht, 2004). In this study, we worked on RFN, RSE, PFN, and PSE pork meats.

Statistical data show that exudative pork meats can cause an economic loss of $5 per carcass (Murray, 2000). Another survey showed that only 16% of the carcasses have ideal lean quality based on color, firmness and water holding capacity (Kauffman et al., 1992). So, the identification of exudative pork meats is a must for the pork industry. PFN and RSE have been recognized recently as major quality defects in Canada. PFN and RSE meats represent more than 13 % of all defects comparing to PSE (13%) and DFD (10%) (Murray, 2000). According to one of the major Canadian pork processor, the RSE incidence may reach as high as 30% (Fournaise and Davies, 2003). There must be an efficient and effective quality assessment system to identify the defects.

The main quality problems in pork meat industry today are “poor colour” and “inadequate water holding capacity”. These two qualities influence the appearance and attractiveness of pork to consumers (Cannon et al., 1995).

Fresh pork colour is visually evaluated by using either the Japanese Pork Color Standards (JPCS) (Nakai et al., 1973) or the National Pork Producers Council (NPPC) Pork Standards as a reference (NPPC, 1996). Although useful, visual evaluation of meat colour can vary with evaluator and may be quite expensive. More objectively, colour can be measured by using a Minolta Chromameter. This instrument measures brightness (L^* value), redness (a^* value), and yellowness (b^* value) of the samples (CIE, 1978).

Drip loss from fresh pork is a result of shrinkage of muscle proteins, especially actin and myosin proteins, and the following expressing of fluids from the meat.

Traditional methods for measuring drip loss are filter paper press method, filter paper method, bag method, and tray method. Although measuring techniques have been practiced for years, an international standard procedure is not available (Otto et al., 2004).

During postmortem, pH declines, cell membranes are disrupted and the amount of intracellular and extracellular fluid changes. So, it would be reasonable to see the change in the dielectric property and connect to the pork meat quality classification (Otto et al., 2004; Fortin and Raymond, 1988: Warriss et al., 1991).

This study was designed to evaluate pork meat quality based on water holding capacity methods, colour values, and vis-spectral measurements.

1.1 GENERAL OBJECTIVES

This study was carried out in two stages. The first stage was comparison of water-holding capacity methods and study of CIE *L*, a*, b** values of four different pork meat classes. The second stage was visible spectral measurements of the four pork meat groups. The specific objectives were:

1. To compare different WHC measuring methods for classification of pre-determined pork meat quality.
2. To investigate CIE *L*, a*, b** values of pork meat for discriminating different pork meat classes.
3. To investigate the potential of using visible spectroscopy for the classification purposes.

CHAPTER 2: GENERAL LITERATURE REVIEW

2.1 MEAT PRODUCTION AND CONSUMPTION

Meat is the main protein supplier in human diet. Meat can be classified as red meat and white meat based on its colour. Cattle, sheep and pigs are "red meat species" and poultry is "white meat species". Beef is the most important red meat species in North and South America, Africa, and Europe whereas sheep are the most important in the Near East and pigs in the Far East (Warris, 2000).

Although it appears that beef has been most common, this trend has changed in 10 years. It is reported that the total world production of the four main types of meat (cattle, sheep, pig and poultry) in 1995 was 197 MT (metric tons) (Warris, 2000). The largest amount of this production was pig meat by 83.2 MT and followed by beef 53.2 MT, sheep meat 7.0 MT and poultry 3.9 MT. First of all, pork is cheaper to produce than beef and sheep. Secondly, in most of the developed countries pigs and poultry are thought to be healthier to eat. Another factor is that in some parts of the world pork and poultry are traditionally preferred meats. For instance, in the years between 1984 -1994, pork production increased by 122% in China, whereas in the European Union (EU) the increase was 45%, and in USA it was only 20% (USDA, 2006).

2.1.1 Canadian pork meat production

Canada is the third most important export market for the US according to USDA International Meat Review, Nov.2006 release. It was estimated that Canadian pork production during 2006 declined 1.5% below the level of the previous year. There has been a significant hog production loss in Ontario and Quebec because of porcine circovirus. Also, anticipation about increased U.S. pork output may be putting downward pressure on Canadian hog market prices resulting in additional production decline for Canadian pork in the coming years (USDA, 2006).

2.1.2 Nutritional aspects of meat

Meat is an important source of protein, vitamin B1, niacin, B2, B6 and B12 and vitamin A. It is also a major source for iron, copper, zinc, and selenium. When all animal products are included whole in protein supply for humans, red meat and poultry make up one sixth of all protein consumption.

Meat generally contains relatively high fat content. This fat is mostly separable fat. But that does not mean the lean part has no fat. It still contains 1 or 2% fat. For instance white (breast) meat of poultry contains almost 1% fat. For poultry, the fat content increases if the skin is counted. Skin contains almost 33% fat. The fat content of skin is much higher if it is taken from subdermal layers (Warris, 2000).

Nutritive value is basic to high pork quality. Pork meat contains a desirable combination of essential amino acids in a biologically available form, the water-soluble vitamins, especially thiamine, some minerals, notably iron, and high-energy lipids including essential fatty acids (Meat Evaluation Handbook, 2001).

For pork, recommended intramuscular fat (IMF) content for acceptable palatability ranges from 2 to 4% (Verbeke et al., 1999). Although, intramuscular lipids play an important role in the sensory and biophysical properties of pork meat, there is still much debate concerning health aspect. While marbling or the intermingling of fat with lean has been equated with palatability and tenderness for years, increasing concerns regarding animal fat in the diet has caused the perceived health benefit from fat reduction to receive greater importance than assurances of tenderness or palatability. Thus, consumers want minimal visual fat and palatable product, making it difficult to satisfy their requirements. On the other hand, consumers have clearly showed a preference for intramuscular fat when rating pork in blind taste tests (NPPC, 1996).

2.2 THE CHEMICAL COMPOSITION AND STRUCTURE OF MEAT

An animal's body consists of chemical substances. It is about 55-60% water. The inorganic component is composed of water plus 3-4% minerals. The remaining part, 35-40%, consists of organic substances. The organic substances are carbon, hydrogen, and oxygen, sometimes with nitrogen, sulphur or other elements. The important three major

organic compounds are proteins, fats and carbohydrates. The approximate chemical composition of pork meat is given in Table 1.

Table 2.1 The Approximate Chemical Composition of Pork Meat (Warris, 2000).

Substance	Percentage
Inorganic	
Water	*60*
Minerals	*4*
Organic	
Proteins	*20*
Fats (lipids)	*15*
Carbohydrates	*1*

There are few scientific reports comparing the chemical composition of chops from different pork meat quality classes. Lawrie (1960), and Wismer-Pedersen and Briskey (1961) found no significant differences between moisture and protein contents of Pale, Soft, Exudative (PSE) pork and normal (RFN) pork meat.

2.2.1 Muscles and their structure

The meat sold in the market is based on skeletal muscle. A muscle is usually enclosed by a thick sheath of connective tissue, the epimysium (Figure 2.1a), and divided into bundles of fibres by a connective tissue network, the perimysium. The individual muscle fibres are surrounded by a plasma membrane itself bound by a thin connective tissue network, the endomysium. This consists of a base membrane surrounded by a reticular layer, in which collagen fibrils are set in a matrix.

The skeletal muscle fibres show very regular crossways striations along their length and the structural unit that is repeated between successive Z-line is called the sacromere (Figure 2.1b). The striations of the myofibrils are caused by a highly organized array of two kinds of filaments: the thick filaments and the thick filaments (Warris, 2000).

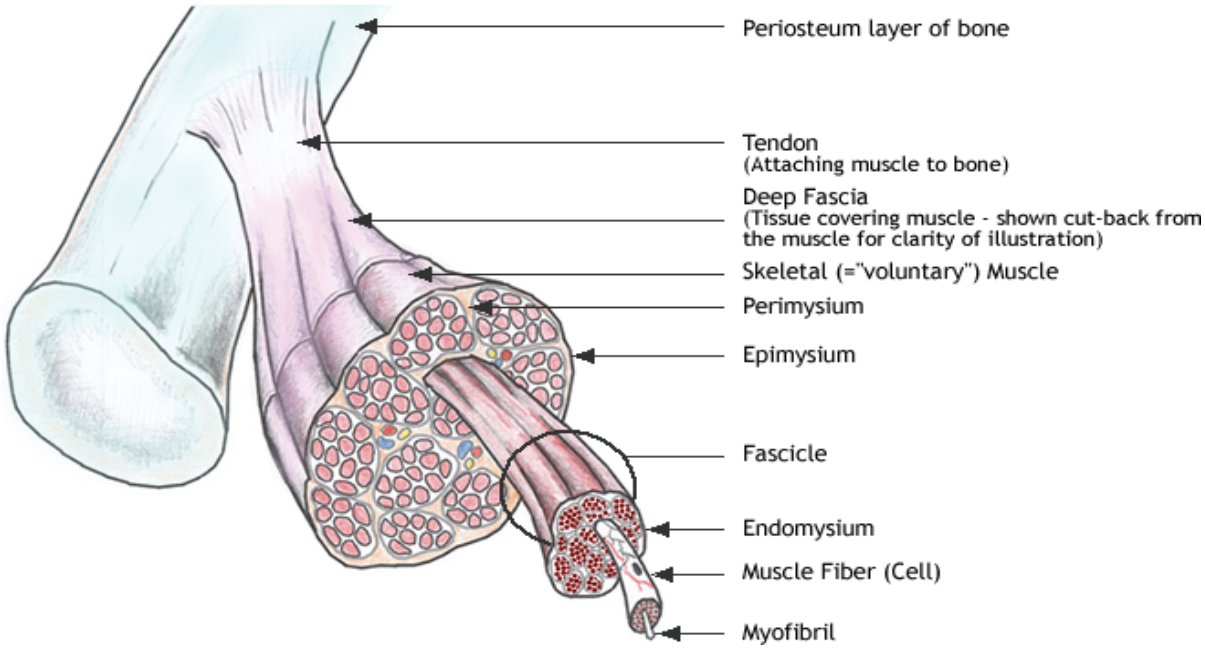

Figure 2.1a Diagram of the structure of muscle and associated connective tissues (http://www.ivy-rose.co.uk, viewed Aug. 29th , 2007)

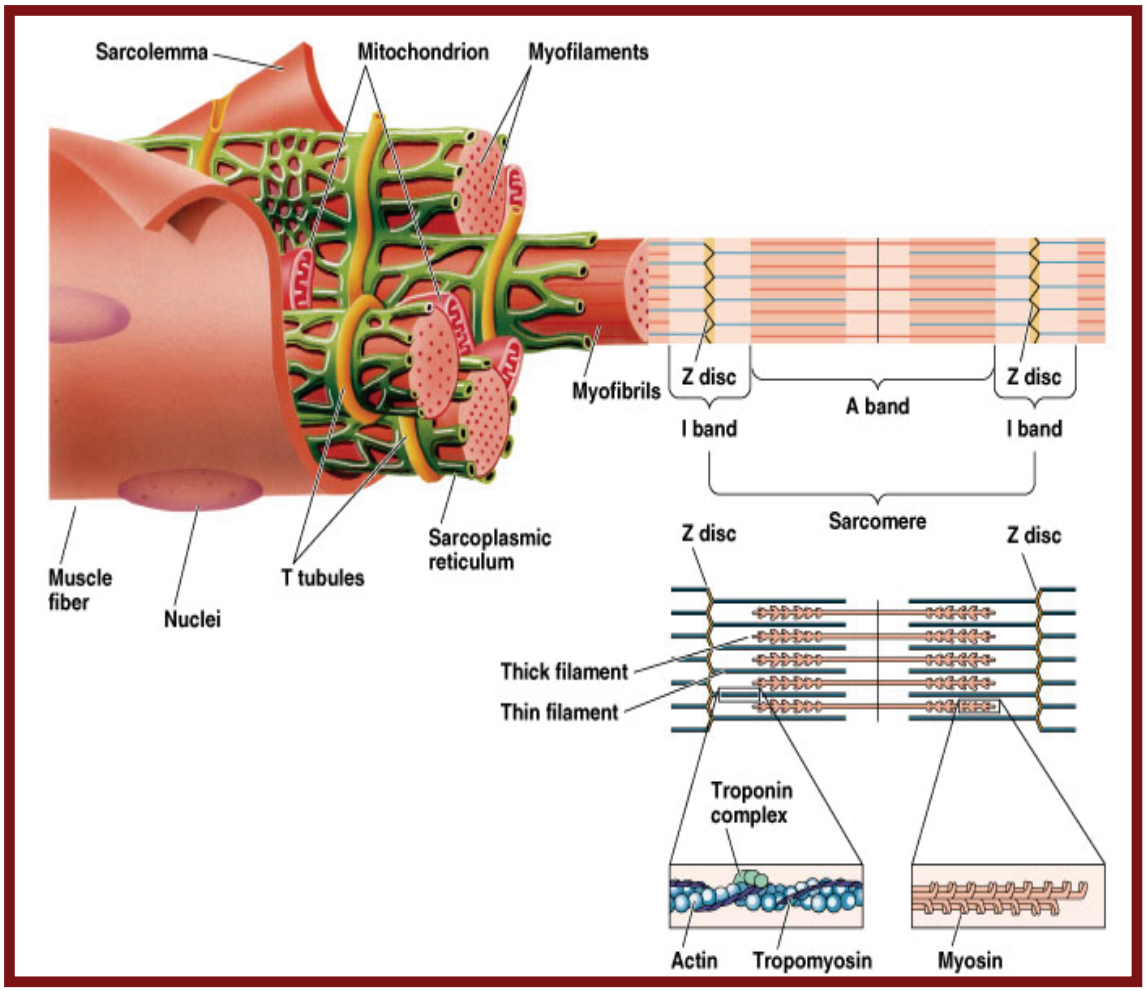

Figure 2.1b The fibrous microstructure of meat
(http://comenius.susqu.edu/bi/320/L5%20Muscle.ppt, viewed Nov. 20th , 2007)

The sliding of myofilaments relative to each other occurs in muscle from living animals with the contraction initiated by the nervous system. This contraction occurs by the actin and myosin filaments sliding with respect to each other. This activity is initiated by release of calcium from the sarcoplasmic reticulum. Then activated and continued by ATP. Usually as a consequence of death, the muscles are in rigor which means in the presence of calcium and absence of ATP. The depletion of muscle energy stocks leads to rigor mortis and a change in status i.e. the muscles becomes meat (Warris, 2000).

2.2.2 Some of the important muscles

Generally, muscles'names describes the muscle position or characteristics. For instance, *M. longissimus dorsi* (*Musculus longissimus dorsi*-LD) (Beecher et al., 1965), sometimes named *as M. longissimus thoracis et lumborum*, refers to the whole length of the back and the main muscle when it is used for 'chops' or 'rib-steaks' cut from posterior rib region and the loin. Sometimes it is used for 'eye' muscle.

Another example is the *M. psoas* major (PM) or psoas muscle from the 'fillet'. This muscle is from posterior under the transverse processes of the vertebrae in the loin region from the level of the head of the last rib.

Most studies in the literature use the *M. longissimus dorsi* muscle for research (Pedersen et al., 2003; Brondum et al., 2000; van Oeckel et al., 1999 and Kauffman et al., 1986a).

2.3 MEAT QUALITY

Having better meat quality is the basic concern of all meat producer and processor. Many factors affect the total quality of fresh and processed meat products.

Every consumer or producer in any industry wants or expects an optimal quality and price and consistency when they purchase something. This is a requirement for producers when they buy raw material and consumers also when they shop. This situation has led to certain requirements from the meat industry. Meat must have, first and foremost, technological quality; secondly the meat industry must guarantee the meat's safety and finally its authenticity (Monin, 1998). Technological quality is a term used for describing meat for further processing like salting, curing, storage, etc.

In the following sections, the definition of meat quality and some major factors influencing the quality will be summarized.

2.3.1 Meat quality definition

There is no standard definition of meat quality that meets all the quality components of the meat production. Health and ethical properties could be as important as technological and sensory characteristics of meat. All of them form some part of the definition with varying importance to what it is called "meat quality". For consumers,

criteria such as uniform colour, little visible fat, a high water holding capacity will be very important to decide to purchase fresh meat. Also eating quality will be as important as physical appearance of the meat at the time of decision to buy. For processors, fundamental factors like pH in combination with the capacity to take up water or, just the opposite, to lose water will be crucial. For instance, these parameters change for dry cured ham production as opposed to the production of cooked ham. So, the definition of meat quality includes different factors according to purchaser's or producer's demands. Key factors of most quality assurance schemes are food safety and ethical aspects. Also, sensory and technological aspects deserve more attention due to the demands of consumers and producers (Hambrecht, 2004).

Another factor should be mentioned here is that differences between culture and people. For instance, in an interesting experiment carried out to understand the cultural effect on quality perception, Spanish and British taste panels examined the eating quality of meat from lambs produced in Spain and the UK. Both panels agreed that the best flavor was from the British lamb and that the Spanish meat had a higher juiciness. But when it came time to ask which meat they would prefer, the British panel chose the British lamb and the Spanish panel chose the Spanish lamb. This shows that meat quality preferences can be influenced by previous experience and conditioning, and may be quite different for different people/cultures (Sanudo et al., 1998).

2.3.2 The major factors affecting quality

A complete list of quality characteristics is given in the following Table 2.2.

Table 2.2 The Major Components of Meat Quality (Warris *et al.*, 1996).

Yield and gross composition:	Quantity of saleable product, Ratio of fat to lean, Muscle size and shape,
Appearance and technological characteristics:	Fat texture and color, Amount of marbling in lean (intramuscular fat), Color and WHC of lean, Chemical composition of lean
Palatability:	Texture and tenderness, Juiciness, Flavour,
Wholesomeness:	Nutritional quality, Chemical safety, Microbiological safety,
Ethical quality:	Acceptable husbandry of animals

Each component will be discussed individually in the following sections.

2.3.2.1 Yield and Composition

Yield refers to how much of a product is sold. Therefore, the more products available to sell, the higher the potential profit to gain. In the meat industry, a larger muscle to bone ratio will result in greater profit for the producer. However, composition is also important. Having a higher ratio of muscle to fat is the main goal for the European and North American consumers. At the same time minimum fat is required because of the taste issue concern (Warris, 2000).

2.3.2.2 Appearance and Technological Characteristics

Generally consumers decide to purchase meat based on its appearance. The colour of the meat greatly affects its saleability. Also, its water-holding capacity (WHC) is also important to the consumer. It can be said that appearance and technological characteristics are connected. The importance of WHC can be classified into three sections; firstly, poor WHC can be connected to the appearance of the meat. WHC is obvious to the consumer when examining the Styrofoam packaging in the retail stores. Poor WHC results in the drip remaining in the package - resulting in a negative appearance of the meat. Secondly, the drip loss is connected to the weight of the meat. In processed meats, poor WHC may reduce water retention and therefore yield of product is reduced. Finally, the juiciness of the meat after cooking is also affected by the WHC. Poor WHC meat may be dry or taste may be negatively affected.

Beside colour and WHC, there is also a relationship between appearance and intramuscular fat (IMF) or marbling. This is also an important factor for determining appearance of the meat. High marbling is a requirement for some consumers such as in Japan, whereas low marbling is required by some other countries as in France (Monin, 1998; Warris, 2000)

Additional detailed information about WHC and colour are found in sections 2.4 and 2.6 respectively.

2.3.2.3 Palatability

Palatability or eating quality of meat can be defined by three characteristics. Those are tenderness, juiciness, and flavour or odour. In most countries, people want their meat tender, but that is not the case for many African countries, where they prefer their meat chewy.

Juiciness of the meat is mainly related to the WHC of the meat or low IMF level. Flavour and odour are closely related. Generally flavour is linked to water-soluble materials, and odour is related to fat-soluble volatile elements. If the meat smells unpleasant, it is mostly related to the quality of the meat. It can be an indicator of the spoilage. But it is not always the case. For instance, some unpleasant smell can be caused by the boar's taint of male pigs (Warris, 2000).

2.3.2.4 Wholesomeness

According to the Canadian Food Inspection Agency, *"Wholesomeness*" is defined as free from decomposition, bacteria of public health significance or substances toxic or aesthetically offensive to man.

Wholesomeness has two components. First, meat should be safe to eat. This means the meat must be free from parasites, microbiological pathogens and hazardous chemicals (Heitzman, 1996).

Second, people want meat to be beneficial to their health in contributing minerals, vitamins, high value protein, and possibly essential fatty acids, such as eicosapentaenoic (EPA) and docosahexaenoic (DHA) acids to their diet (McCance and Widdowson, 1997).

2.3.2.5 Ethical quality

The last major component of meat quality is the ethical quality. Many people have concern about the meat they consume. They believe meat should come from animals that have been bred, reared, handled and slaughtered in ways that promote their welfare and in systems that are sustainable and environmentally friendly. In other words, this system should be sympathetic to animal welfare.

The Canada Pork Council (CPC) represents Canadian hog producers. CPC has gathered hog producers, animal care scientists and regulators to define good husbandry practices (Forian, 2006)

2.4 PORK MEAT QUALITY

There is no single definition of high quality meat used in the pork industry today. Meat quality is a combination of subjective and objective measurements that vary across markets. Colour, pH, water holding capacity, firmness, and marbling are some of the most common measurements used in determining pork quality (PIC, 2003).

Desirable or undesirable muscle quality is related with the morphological, chemical, physical, biochemical, microbial, sensory, technological, hygienic, nutritional, and culinary properties of the meat. During the conversion of muscle to meat, the rate of glycogen conversion to lactic acid as well as the accumulated lactic acid is very important. This affects the ultimate colour and water-holding capacity of the muscle.

Deviation from the desired conversion of muscle to meat can cause the production of three major problems in pork quality: Pale, Soft, and Exudative (PSE), Red, Soft, Exudative (RSE) and Dark, Firm, and Dry (DFD) pork (Meat Evaluation Handbook, 2001).

The ideal method for assessing pork quality is by the direct evaluation of the *longissimus muscle* and the back fat in the same general area. Colour, wetness, firmness, texture and marbling content of the exposed loin eye are the primary lean quality characters. For subjective evaluation of pork meat, National Pork Producers Council developed colour, wetness/firmness, and marbling standards as shown in Figure 2.2.

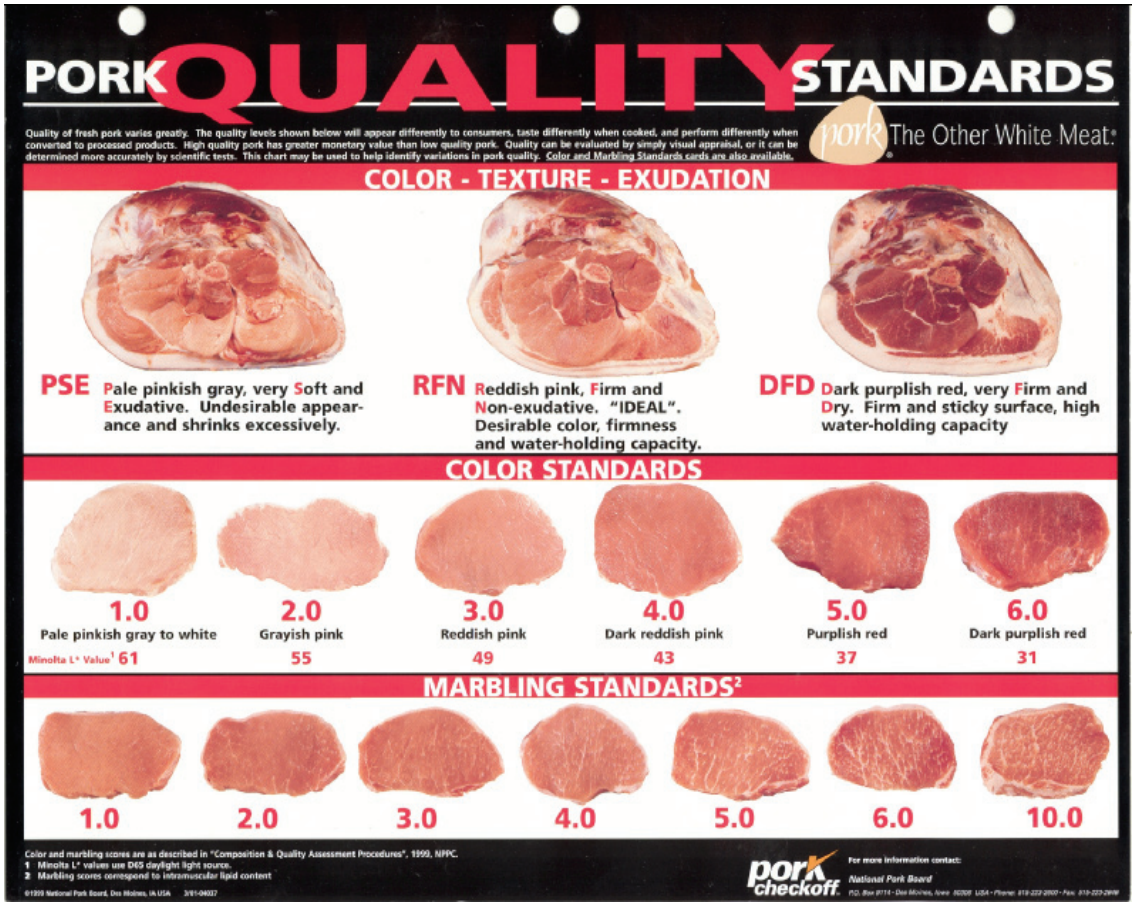

Figure 2.2 Pork Quality Standards (NPPC, 1999)

Two of the most used measurements, colour and water-holding capacity (WHC), will be discussed in detail in the following sections. However, before discussing colour and WHC, a discussion about some of the critical control points in pork production affecting the quality should be mentioned also. Those critical control points are genetic input, nutrition, on-farm handling, transport, preslaughter treatment, stunning, early postmortem handling of carcasses, chilling, further processing and preparation by the consumer. Only brief information will be given about genetic input, nutrition and transport and preslaughtering treatment.

2.4.1 Genetic input

The effect of breed is significant for many quality characters such as WHC, pH or intermuscular fat. Meat from Pietrain pigs generally exhibits the PSE condition because of presence of the Halothene gene, which causes high stress susceptibility. This gene causes increase in metabolism, for example, intense production of heat and lactate and contraction of skeletal muscles. Pigs carrying this gene show a higher muscle temperature both ante- and post-mortem and a more rapid pH-decline post-mortem due to the increased turnover of glycogen to lactate. Even if it is a recessive gene, carriers of the gene still tend to have a worse meat quality than non-carriers (Oliver et al., 1993; Sellier, 1998).

Another problem is caused by the RN- gene. This gene is frequently found in meat from Hampshire pigs. Those pigs show a very low water holding capacity because of the RN- gene, which is frequently encountered in that population. It was suggested by Warner et al. (1997) that this gene might be responsible for the development of RSE meat. However, van Laack and Kaufmann (1999) demonstrated that RSE meat samples showed a normal glycolytic potential. In another words, RSE meat is not the result of presence of the RN- gene but rather a mild form of PSE meat.

2.4.2 Nutrition

Nutrition may affect meat quality by means of feeding level and feed composition. It is said that a higher feeding level has positive effects on tenderness and juiciness of the meat (Wood et al., 1994). Also the ingredients making up the feed have

crucial effects on the quality. For instance, effects of dietary fat composition on the fatty acid profile in both the intramuscular fat and other fat depots (Warnants et al., 1999). Fatty acid composition of the phospholipids fraction of the intramuscular fat could have an effect on membrane stability, oxidation processes, flavour development and possibly water holding capacity (Monahan et al., 1992; van Laack and Spencer, 1999).

Another example of feed ingredients can be given such as vitamin E and selenium supplementation. They may improve dressing percentage, reduce lipid oxidation, and increase alfa-tocopherol concentration of tissues (Corino et al., 1999).

2.4.3 Transport and pre-slaughter treatment

This might be the most important control point for meat quality control. Transportation conditions such as noise, loading and unloading fighting due to the mixing of unfamiliar pigs and stocking too many animals in a truck mean severe stress for the animal resulting in an accelerated post-mortem glycolysis and impaired meat quality (Smulders and van Laack, 1991). Ideal stocking density should be spacious enough to allow the pigs to rest and yet not too spacious to prevent pigs from losing their balance and fighting between themselves. Optimal stocking density depends on transportation time, genotype and climate. In general, environmental temperature of approximately 16 ºC with a low air velocity is suggested for best meat quality.

After transportation a short lairage time of approximately 2 hours at the slaughterhouse allows the animal to recover from transport stress and may improve both animal welfare and meat quality (Warris, 1987; Maribo, 1998). During transportation and lairage, mixing of unfamiliar pigs should be avoided because fighting and social stress can cause more PSE and DFD pork meat quality (Karlsson and Lundstrom, 1992).

In hot season, showering the animals regularly during lairage could have a beneficial effect against aggressive behaviour to improve the welfare of the animals. Also showering before slaughtering can decrease muscle temperature and may lead to a better meat quality.

2.5 WATER HOLDING CAPACITY

The meat quality can be expressed by several quality characteristics such as flavour and nutritional value. Hoffman (1973) defined four groups of quality classes - eating quality, nutritional quality, technological quality and hygienic quality - to define meat quality objectively. Meat quality generally is described as the sum of all meat quality characteristics (Hoffmann, 1986). Water holding capacity measured as drip loss has high importance in pig meat production because of its' financial implications. In general it can be said that meat with a high drip loss has an unattractive appearance and this leads to loss of sales (Otto et al., 2004). According to Kauffman et al. (1992), it is reported that unacceptably high moisture loss from fresh product has been estimated to occur in as much as 50% of the pork produced.

Water holding capacity, or WHC, can be described in several ways. Water-holding capacity is often described as drip loss or purge. Water-holding capacity of meat is described as the ability of the post-mortem muscle to retain water even though external pressures are applied to it. Muscle contains approximately 75% water, 20% protein, 5% lipids or fat, 1 % carbohydrates, and 1 % vitamins and minerals. Water in muscle cells is a dipolar molecule and is attracted to charged particles like proteins.

Most of the water in muscle is held within the myofibrils, between the myofibrils themselves and between the cell membrane (sarcolemma), between muscle cells and between muscle bundles (groups of muscle cells). After the muscle is harvested, the amount of water in meat can change according to many factors depending on the tissue itself and how the product is handled. The water in muscle has many different forms including bound water, entrapped water, and free water. Each of these forms of water will be discussed in further detail below (NPPC, 2002).

2.5.1 Important definitions for the water-holding capacity of fresh meat

2.5.1.1 Bound water

Water is a dipolar molecule which is attracted to particles species like proteins. Actually, some of the water in muscle cells is closely bound to protein. Bound water is the water that exists in the vicinity of non-aqueous constituents (like proteins) and has

reduced mobility, in another words, it does not easily move to other compartments. This water is very resistant to freezing and to being driven off by conventional heating. True bound water is actually a very small fraction of the total water in muscle cells and approximately 0.5% of the water in muscle is truly bound water. Bound water amount changes very little if at all in post-rigor muscle (Fennema, 1985).

2.5.1.2 Entrapped water

Entrapped water is another fraction of water that can be found in muscles and the water molecule in this fraction can be held either by steric effects and /or by attraction to the bound water. In early postmortem tissue, this entrapped water does not flow freely from the tissue; however it can be removed by drying, and can be easily converted to ice during freezing. Entrapped water is affected by the rigor process and the conversion of muscle to meat. Because of changing in muscle cell structure and lowering of the pH, this water can eventually escape as drip loss.

The entrapped water in living muscles makes up 80% of the total water in muscle. During the conversion of muscle to meat, mostly the entrapped water is mostly affected. So maintaining this water is the main goal of many processors (Fennema, 1985). Some of the factors that affect the retention of this water include net charge of myofibriller proteins and the structure of the muscle cells and its components, as well as the amount of the extra-cellular space within the muscle itself (NPPC, 2002).

2.5.1.3 Free water

It is defined as the water whose flow from the tissue is unimpeded. Mainly, weak surface forces hold this fraction of water in meat. Generally free water is not seen in pre-rigor meat. However, it can develop as conditions change that allow the entrapped water to move from the structures where it is found (NPPC, 2000).

2.5.2 Factors affecting drip

2.5.2.1 Genetics and early postmortem handling

In some cases, genetics and the handling of the live animal can play a big role in influencing the future water-holding capacity of that product. Also, the way of handling, particularly with respect to cooling when it enters the rigor, plays a critical role in influencing the amount of moisture that will remain in the product. Each of these factors - genetic, live animal handling and early postmortem handling - has the potential to greatly influence pH decline, and, as a result, the water-holding capacity of the meat. Thus, it is crucial for all levels of the industry to understand how these factors can interact to affect water-holding capacity (NPPC, 2002; Gardner, 2004).

2.5.2.2 Rate of pH decline

pH is most commonly measured in fresh meat because it affects technological ability, and most sensory traits. To illustrate, in France, ultimate pH measurements have been practiced in pig selection since 1981 (Monin, 1998).

Low ultimate pH and accelerated pH decline are related to the development of low water-holding capacity and, consequently, high purge loss. The rapid pH decline causes the denaturation of many proteins. The most severe drip loss is seen often in PSE meats, which are from pigs that have inherited a mutation in the ryanodine receptor/calcium release channel (halothane gene) in the sarcoplasmatic reticulum. This release of calcium causes rapid contraction and then increases in the rate of pH decline. This mutation in the halothane gene can be recognized in the parent stock. In the United States Pork Industry, this gene is almost eliminated in most commercial herds because a commercial test for this mutation exists. Of course the halothane gene is not the only reason why severe drip loss occurs in PSE meat. For instance, before harvest, even for normal animals short-term stress can accelerate metabolism thus causing a more rapid pH decline. Besides, it should be noted that although the pH of these muscles falls faster than normal, the ultimate pH may not be below normal ranges (NPPC, 2002).

It should be noted that pH is generally measured within one hour of slaughter (initial pH) or within 24 hours (ultimate pH or pHu) and then stays stabile (PIC, 2003).

2.5.2.3 Pre-rigor temperature

The denaturation of proteins is not caused only by pH decline. It is the combination of relatively acidic conditions and also body temperature. So it is advised that rapidly chilling the temperature will reduce some of the effects of lower pH. In addition, lowering the temperature also decreases metabolic processes and reduces the rate of pH decline. So using early and intensive chilling can prevent mild cases of PSE. Actually in severe PSE, the rapid pH decline can make it difficult to lower the muscle temperature fast enough to prevent protein denaturation (NPPC, 2002).

2.5.2.4 Processing factors

In addition to live animal management and early postmortem handling of the meat, some other factors including storage time, physical disruption of the product and storage conditions also have an effect on water retention.

In pre-rigor meat, very little drip loss occurs. At later times after slaughter, when the muscle has gone into rigor, drip losses tend to increase (Jolley et al., 1981). One reason could be that formation of rigor bonds decreases the amount of space, which is available for water to occupy in the myofibril. Another reason for this is that the pH of the tissue is close to the isoelectric point of many of the major proteins (especially myosin). Therefore, this affects the amount of water, which is attracted to protein structures in the myofibril. As a result of those two factors, the amount of drip loss could increase (Offer et al., 1989).

Number of cuts and size of the pieces affect the percentage of the product that is lost as drip. Smaller cuts cause more drip loss than do larger cuts.

Another processing factor which affects the purge is storage conditions. For instance, increasing the temperature from 0 to 4 °C can cause an important increase in drip loss (Sayra et al., 1964).

Freezing and thawing of fresh meat are also important and affect drip loss. Penny (1975) reported that frozen and thawed pork can have almost a two-fold increase in drip loss compared to non-frozen pork. It is because of the physical disruption caused by ice crystals formed in the meat. If the meat is frozen very quickly, then the meat quality is better because fast freezing forms small crystals. While slow freezing causes

bigger ice crystals to form and it results in expansion and even rupture of the cell membrane. As a result of slow freezing, significant damage to the cell membrane occurs (Love and Haraldsson, 1961).

2.5.3 Measuring methods of WHC of pork meat

There are many available methods that have been used to measure WHC. Unlike colour, WHC is not definable in absolute units since each method measures slightly different things. A comparison of methods used to measure WHC in pork was given by Kauffman et al. (1986a and b), and a general overview of WHC methods by Honikel and Hamm (1994). Also, comprehensive reviews of WHC are given in Offer and Knight (1988a and 1988b) and Offer et al. (1989).

Traditional methods of measuring WHC are the filter paper press method first introduced by Grau and Hamm (1953), the filter paper method (Kauffman et al., 1986a and b; Hamm, 1986) the bag method (Honikel, 1987), and the tray method described by Lundstrom and Malmfors (1985). Additional methods used to measure drip loss are described by Honikel and Hamm (1994). Recent studies show that there is need to find more suitable methods for determination of drip loss. Rasmussen and Andersson (1996) recommend a method working with EZ-DripLoss containers. Another group of researchers, Walukonis, Morgan, Gerrard, and Forrest (2002) used absorptive material in the early post mortem stage. However, there is high diversity in procedures such as in sample size, or the force applied to the meat. For instance, bag method is carried out with approximately 100 g samples, whereas the EZ-DripLoss method uses approximately 10 g and for centrifugation almost 3-4 g.

Based on the force applied to the meat, the methods can be distinguished into gravimetric methods (bag, EZ-DripLoss, tray method), absorptive or capillary action methods (filter paper methods, cotton-rayon material method) and methods where external pressure is applied to the meat as in the filter paper press method and centrifuge method. However, because of the variation in the methods used, the results for drip loss in the literature are difficult to compare (Honikel, 1998).

WHC can be evaluated using three different methodologies - gravimetric methods, absorbent methods, and enhanced force methods (methods where external pressure is applied) (Warris, 2000).

2.5.3.1 Gravimetric methods

This method is the simplest method. In this method sample muscles are stored for a known period of time and the loss of drip loss is measured. It is common to suspend slices of the loin inside polythene bags to prevent evaporative losses and the samples are kept at 1 to 5 ºC for 48-72 hr, sometimes even for 192 hr (Wilborn et al., 2004). The most commonly used gravimetric procedure – the bag method - was proposed by Honikel (1987). Although this method has gained international acknowledgment, it is quite space consuming (Christensen, 2003). In this method, *M. longisimuss dorsi* form which associated fat is removed is suspended by a thread or in a plastic net and enclosed in a sealed plastic bag at 0-4 ºC. The sample muscle is weighed before the hanging. It is re-weighed after storage for 48 hr or longer. In the study of Kauffman et al. (1986a), it was reported that the 48 hr drip loss was used rather than 96 hr because it was determined that this is the most practical duration in which fresh pork cuts are stored, transported or displayed. Also, this is the standard time used as a measure of percent drip loss in suggested research tests (Honikel, 1985). It should be noted that if making measurements on the m.longissimus dorsi muscle, it is important to understand that WHC can vary in different parts of the muscle.

In addition to the bag method, there are several applications that use the gravimetric methodology. Some of the other applications are EZ-DripLoss (Rasmussen and Andersson, 1996), tray method and Danish drip tube method developed by Rasmussen and Andersson (1996). From those methods, the EZ-DripLoss method has the ability to detect local PSE spots (Christensen, 2003). This method is a new procedure recommended by the Danish Meat Research Institute (DMRI) for routine measurements of drip loss (DL) in pork meat. (Rasmussen and Anderson, 1996; Christensen, 2003).

Current methods used to estimate water-holding capacity of postmortem muscle by measuring drip loss are time-consuming (Offer and Trinick, 1983). It was reported

that drip loss methods, especially when size is standardized, are quite appropriate if time required to obtain results and field application are not important (Kauffman et al., 1986a).

2.5.3.2 Absorbent methods

Capillary action or absorbent methods include the use of absorbent materials such as filter paper, or cotton-rayon material. All of the gravimetric methods require that muscle samples be removed at 24 hr post mortem (PM) and allowed to drip for a given period of time, typically 24 hrs or 48 hrs. Walukonis et al. (2002) suggested that using an absorptive material during the early postmortem period may be a useful and accurate way of predicting WHC. In this study cotton-rayon material (~2 g; o.b. Regular Absorbency Tampons, Johnson&Johnson, NJ) was inserted in the longissumus muscle through the subcutaneous fat layer. Absorbance was calculated as the difference between the final weight and initial weight of the material.

Filter paper technique is cheap, easy and very fast and does not need special equipment, certainly for visual scoring (Van der Wal et al., 1988). For rapid, inexpensive and accurate results, the filter paper tests proved worthy of consideration and are comparable in accuracy with the other test (i.e. drip loss and filter paper press methods) (Kauffman et al., 1986b). In this method, a dry, pre-weighed filter paper is pressed lightly onto the surface of sample, removed after 3 seconds, and weighed again. This is performed after a 10-20 minute waiting period for the freshly cut chop. The absorbed moisture is calculated as the difference between weights (Chan et al., 2002).

2.5.3.3 Enhanced force method

Enhanced force methods are filter paper press and centrifuge methods. The filter press method was originally developed by Grau and Hamm (1953). A small piece of meat sample (approximately 0.2-0.4g) is pressed on a filter paper between two clear plastic plates to form a thin film. Because of applied pressure, water is squeezed out and absorbed by the filter paper and a ring of expressed juice forms. The ratio area of this ring to the meat is an index of WHC. There will be a larger area for the meat with a high WHC than the meat with a low WHC. The main advantage of this technique is that it can be employed with ground or processed meat. Also, the sample size is small and

operation is easy. The main disadvantage is that the measurements of meat and liquid areas made with the planimeter are inefficient, laborious and unstable (Irie et al., 1996). In the study of Kauffman et al. (1986b), the filter paper press methods proved to be quite disappointing, especially since they have been used longer in meat research than most other techniques studied. It is likely that excessive evaporation while standardizing sample weight, small sample size, difficulty in applying uniform and constant pressure, and difficulty in assessing a reliable area measurement when using the planimeter, all combine to produce erroneous and poor measurements of WHC.

For the centrifugation method, small samples of meat are centrifuged at high speed for a certain time. The weight differences between the sample before and after centrifugation are recorded (Honikel, 1998). It was recorded that centrifugation methods serve as reliable laboratory procedures but require more initial investments of equipment and time (Kauffman et al., 1986a and b).

2.6 PORK COLOR AND APPEARANCE

"Pork: the other white meat ®" (NPB, 2000), is the fifth most familiar advertising slogan in the USA. In fact, this slogan was a play on words that repositioned pork as a white meat to focus on its nutritional value. In spite of the success of proclaiming pork as a white meat, fresh pork colour that is comparable to poultry has been linked to extremely undesirable pork quality. When consumers buy fresh pork based on a visual appearance of "white colour", they may fail to select a quality muscle food (Norman et al., 2003).

Colour plays a major role in consumer evaluation to purchase and bright red colour is connected to freshness. Thus, colour may be the most important factor that influences the appearance and attractiveness of pork to consumers (Faustman and Cassens, 1990). Although colour has a special significance among the pork quality attributes, unfortunately it is difficult to assess because the colour of a meat cut, even within the same muscle, is frequently not uniform (AMSA, 1991).

In food engineering research, it is often necessary to analyze the surface colour of food samples both qualitatively and quantitatively. Primarily a visual inspection and comparison of the food samples is performed. A second analysis may involve obtaining colour distribution and averages (Yam and Papadakis, 2004).

2.6.1 Meat colour

When an oscillating electron drops from a higher energy state to a lower energy state, energy is emitted (Tinoco et al., 1978). Colour occurs if electromagnetic radiation in the visible range is emitted or reflected by atoms or molecules. In the end, colour is related to the electron structure of the atom or pigment molecule. In-coming energy may be absorbed by these electrons, altering their energy states. The light directed at a material or meat sample contains varying amounts of wavelengths in the visible region (~ 400-700 nm). Pigments absorb some of the wavelengths, while some of the wavelengths are reflected. The light, which is reflected back to the eye, is missing the colour associated with the wavelengths that have been absorbed. The reflected light, which is missing some wavelengths, extracts colour (McDougall, 1983).

Meat colour depends on pigment (myoglobin, hemoglobin) concentration, their chemical states, and the light scattering properties of meat (Lawrie, 2002; McDougall, 1983). Myoglobin is a water-soluble protein that stores oxygen for aerobic metabolism in the muscle. After cutting, meat colour is quite dark. When oxygen from air comes into contact with the exposed meat surfaces, it is absorbed and binds to the iron. The myoglobin is then oxygenated. This pigment is called oxymyoglobin. This is the colour consumers relate with freshness. Myoglobin and oxymyoglobin have the capacity to lose an electron, also called oxidation. If oxidation occurs, then it causes a brown colour and it produces metmyoglobin. So, myoglobin can have colour from a dark purple colour to a bright red colour by losing electrons. The pigments myoglobin, oxymyoglobin and metmyoglobin can change from one to the other depending on the conditions in which the meat is stored. Consequently, the relative proportions of myoglobin forms such as deoxymyoglobin, oxymyoglobin and metmyoglobin affect the colour hue of fresh pork. A bright pink or red colour is related to oxymyoglobin, purple is related to the colour of myoglobin and grayish or brownish pink is related to metmyoglobin (Lindahl et al., 2001).

The perception of colour is very dependent on the observer. It is crucial to know the value of relative objective colour measurements to the subjective judgement of acceptable colour (Van Oeckel et al., 1999).

2.6.2. The factors affecting meat colour

The apparent colour is affected by the amount of water in or on the fresh meat. Proteins in meat with a low pH (<5.4) do not bind water very tightly. This free water in the tissues reflects light in many directions, or scatters it. So, it makes the meat appear very light compared to higher pH meat in which water is more tightly bound. Although these two pieces of meat could be the same colour red, one may appear much lighter (pale, soft exudative) than the other (dark, firm, dry). As a result, consumers may think one is redder than the other.

The colour of red meat can change over time as the pigments bind oxygen then ultimately become oxidized to brown or grey. Color can change because of microbial growth, cooking and exposure to various ingredients, for instance vinegar or salt.

And finally, the presence or absence of oxygen in the surrounding environment will also have an impact on the colour of the meat (Mancini and Hunt, 2005).

2.6.3 Measurements of colour

Traditionally, meat colour is assessed by photometric or spectrophotometric methods (reflectance, CIELAB or chromatic coordinates), using surface spectrophotometers, or internal (fiber optic) measurements. Swatland (1995) has reviewed the principles of colour measurements. Cornforth (1994) researched the factors that determine meat colour and MacDougall (1994) has reviewed the measurement of colour. Portable spectrophotometers are convenient for carcass colour grading in industry (Denoyelle and Jabet, 1997).

During the Pork Chain Quality Audit, packers reported a 10% occurrence of PSE pork and a 4% occurrence of DFD pork (Cannon et al., 1996). The relationship between PSE and DFD quality defects and their respective colours has led the industry to assign visual color scores to the carcass. Variation related to different instruments used to measure the Japanese Pork Colour Standards (JPCS) was evaluated at the National Pork Produces Council (NPPC) Training Session (NPPC, 1996). The L^* a^* b^* model is an international standard for colour measurement. The L^* value represents luminance or lightness component, ranging from 0 to 100. The a^* value is the component from green to red and the b^* value is the component from blue to yellow. The L^* a^* b^* colour is

device independent, in other words, providing consistent colour regardless of the input or output device such as digital camera, scanner monitor and printer (Yam and Papadakis, 2004). CIE *L*, a* and b** values (CIE, 1978) were calculated for a variety of operating conditions after determining the spectral curve for each of the six JPCS. The use of instrumental colour evaluation is significant to the meat industry because of its speed, and consistency.

Presently, fresh pork colour is visually evaluated by using either the JPCS or the NPPC as a reference. Tan et al. (2000) studied the ability of Colour Machine Vision (CMV). Untrained panelist evaluated over 200 pork loin chops using either JPCS or NPPC reference standards. Representative samples were used to train neural-network-based image processing software. Then after training, the CMV system was used for evaluation of pork samples based on color distribution. It was reported that training the CMV system by using images of actual meat samples resulted in a stronger correlation to panel scores than training with either set of artificial colour standards. The results of the study showed that CMV is a rapid and repeatable means of evaluating pork color.

Because of the advantages of being nondestructive, free of chemical preparation and having a fast inspection speed, spectroscopy has been studied extensively for determining properties of agricultural products, but less for meat products as compared to plant materials (Chan et al., 2002). Concerning the application of spectroscopy, especially when using Near Infrared Spectroscopy (NIRS) for prediction of the quality of fresh meat, mostly beef tenderness was studied (Byrne et al., 1998; Hildrum et al., 1995; Liu et al., 2003; Park et al., 1998; Shackelford et al., 2005). There are not many reports for prediction of pork quality using NIRS based models. Actually many quality characteristics are related to visible characteristics such as colour. Consequently, reflectance spectroscopy has recently spanned the NIR range to the visible range (Chan et al., 2002; McCaig et al., 1992; Savenije et al., 2006).

In addition, fiber optics has been employed as a means of performing remote measurements inside muscles on the intact carcass (MacDougall, 1980; Swatland, 1989).

More recently, the usage of hyperspectral colour measurements has increased in food research areas, and Jun et al. (2007) studied hyperspectral imaging techniques for pork quality level assessment. According to this study, feature wavebands images set

selected by PCA (Partial Component Analysis) from the first derivative data, yield the best classified result of 87.5 % correction.

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CONNECTING TEXT

Water holding capacity measured (WHC) as drip loss has importance in pig meat production because of its financial implications. High drip loss can cause unattractive appearance and this can lead to loss of sales. There are several ways to measure WHC of pork meat.

The aim of the following chapter was to compare different WHC measuring methods for grouping pork meat classes. Discriminant procedure was performed to classify different pork meat groups. Also, the colour of pork samples was studied for discriminating pork meat classes.

Part of the work has been presented at the NABEC Conference-2006, in Montreal: *Gunenc, A., J. Qiao, M.O. Ngadi, and N. Wang, 2006. Water Holding Capacity of Pork Meat.*

CHAPTER 3: MEASUREMENT OF WATER HOLDING CAPACITY AND COLOUR OF PORK MEAT CLASSES

3.1 ABSTRACT

This study was designed to compare different methods of measuring water holding capacity (WHC) for classification of pork meat according to four defined quality classes - PFN, PSE, RFN, and RSE. The WHC measuring methods were bag method (2 and 4 days), centrifuge method, filter paper and cotton-rayon material method. Also, CIE *L*, a*, b** values and E (E= *a*/b* +a* /L**) values were studied. Discriminant procedure was performed for classification of pork samples into P–R (Pale-Red) and FN-SE (Firm, Non-exudative–Soft, Exudative) groups. Stepwise procedure was used to choose parameters for classification purposes. We had 83% overall-accuracy when classifying the pork samples into P-R groups based on CIE *L** and *a** colour values. The overall-accuracy for classification into FN-SN grouping was 64% with cotton-rayon material and 61 % with filter paper method. From all WHC methods, cotton-rayon material method could be used for classification into P-R and FN-SE groupings. None of these measurement methods alone was enough to separate all pork samples into 4 quality classes alone.

3.2 INTRODUCTION

The ultimate meat quality is a result of the rate and extent of post mortem changes. Following slaughter, the circulatory system can no longer transport nutrients and oxygen to the muscles or remove waste products or heat. In this case, anaerobic pathways are used for energy production, and lactic acid is produced as a by-product of anaerobic metabolism. The accumulation of lactate causes the pH of the tissue to decrease. This accumulation continues until either glycogen stores are depleted or until the pH is too low for the glycolytic enzymes to function. Severe decline of pH can cause extensive denaturation of many proteins in meat. The rate and the extent of pH decline have major impacts on meat quality, especially water holding capacity (NPPC, 2001).

Water Holding Capacity (WHC) or sometimes called water-binding capacity may be the most important of all main characteristics of pork meat that affect appearance,

shrinkage, processing properties and palatability (Toldra, 2003; Kauffman et al., 1986). WHC is defined as the ability of muscle to retain naturally occurring moisture, and generally expressed as drip loss or purge (NPPC, 2002). The WHC of meat is crucial for two reasons: first meat is sold by weight and any loss is undesirable economically; secondly, the WHC influences the appearance of the meat and subsequently the decision of consumers to purchase it (Martens et al., 1982). Gusse (1996) reported that 2% difference in drip loss (DL) between exudative and normal pork results in a crucial economic loss for the processor. Water exudation can cause $5 loss per carcass (Murray, 2001) and up to 40% unmarketable product (Grandin, 1993). Drip loss from fresh pork is a result of shrinkage of muscle proteins, especially actin and myosin, and the subsequent expressing of fluids from the meat. At low pH, the ability of proteins to bind water is decreased, and this causes drip loss to increase. Also, fast decrease in pH causes the actin-myosin complex to contract and expel more fluid from the meat (Lawrie, 1985).

Based on colour, exudative and firmness, good quality of pork meat is referred to as RFN (Red, Firm, Non-exudative) meat whereas defective meat classes are grouped as DFD (Dark, Firm, Dry), PSE (Pale, Soft, Exudative), RSE (Red, Soft, Exudative) and PFN (Pale, Firm, Non-exudative) meat classes (van Laack et al., 1994; Joo et al., 1995).

There are several methods for measuring WHC of meat. For instance, in Honikel's gravimetric method (which seems to have gained general acceptance), a piece of meat is hung in a net inside a plastic bag for 48 hr, and sometimes up to 8 days (Wilborn et al., 2004) and then the percentage of weight loss to initial weight is expressed as WHC (Honikel, 1998). Another method is filter paper press method (Grau and Hamm, 1953), where the meat juice from a well-defined small amount of meat sample is squeezed out and absorbed by filter paper. One other method is centrifugation (Honikel, 1998). Finally, there is tray method described by Lundstrom and Malmfors (1985). Further studies have shown that efforts to find more suitable methods for determination of drip loss are being pursued. Rasmussen and Andersson (1996) suggested EZ-DripLoss method, and Walukonis et al. (2002) used absorptive material for measuring WHC. While drip loss has been measured for years, there is no internationally adopted procedure. All the methods can be grouped according to the force applied to the meat as follows: gravimetric methods (bag, EZ-DripLoss, tray method), absorptive (or capillary

action methods) (filter paper methods, cotton-rayon material method) and methods where external pressure is applied to the meat as in the filter paper press method and centrifuge method.

Colour is another parameter that affects ultimate meat quality. It may be the most important factor affecting the appearance and the attractiveness of pork meat to consumers (Faustman and Cassens, 1990). Fresh pork colour is visually evaluated using either the Japanese Colour Standards (JPCS) or National Pork Producer Council Pork Quality Standards (NPPC) as a reference (Tan et al., 2000). More objectively, colour can be measured by a Minolta Chromameter. This method measures the brightness (L^* value), the redness (a^* value), and the yellowness (b^* value) of the sample (NCSU, 2001).

The aim of the present study was to compare the different WHC measurement methods namely the bag methods for 2 and 4 day, filter paper, cotton-rayon material and centrifugation methods. The second objective was to investigate other quality attributes such as CIE (L^*, a^*, b^*), and E values of meat samples.

3.3 MATERIALS AND METHODS

3.3.1 Sampling and sample preparation

Pork loins were obtained from a local commercial cutting house, OLYMEL Plant, in Quebec, Canada. The fresh pork samples, from different quality groups of PFN, PSE, RFN and RSE, were selected by a trained employee who had more than 10 years working experience. The samples were brought to the Food Engineering Laboratory at McGill University. The samples were sliced into chops with a thickness of 1 cm and the experiments were conducted at room temperatures between 20 and 22 ºC.

In order to include as much variations as possible into the analysis, the data collection was spanned several months from February to December in 2006. A total set of 180 pork samples were used for WHC measurements, and 120 samples were used for other quality attributes. The samples were all obtained at 24 hr postmortem from the loin-eye area.

3.3.2 Water holding capacity measurements

Drip loss measurements were conducted using the following methods;

3.3.2.1 Bag method

A slice of meat was hung in a plastic bag and allowed to stand for 2 and 4 days at 4 ºC. Drip loss was calculated as the difference in weight before and after hanging and reported as percent drip loss of initial weight (Lundstrom and Malmfors, 1985; Bertram, et al., 2001; Barton-Gade et al., 1994).

3.2.2.2 Capillary action method (filter paper method)

Freshly cut pork meat slices were lightly pressed by a dry, pre-weighted filter paper (Fisher brand, Dia.:12.5 cm, Quantitative Q8). The paper was removed after 3 seconds. The amount of absorbed moisture was calculated by the difference between weights before and after pressing (Chan et al., 2002; Van Laack et al., 1995; NCSU, 2001).

3.2.2.3 Centrifuge method

Approximately 10 g of pork loin sample was placed in a 50 mL centrifuge tube together with an absorbent material, sodium sulphate. The meat samples were centrifuged at 3000 rpm for 15 minutes and at 25 °C by using a centrifuge (IEC Centra-8R centrifuge, USA). Then the meat sample was removed with forceps, dried with absorbent paper and re-weighted to determine the weight loss. WHC was calculated as a percentage of weight loss before and after centrifugation (Swatland and Barbut, 1995; Abdullah and Al-Najdawi, 2005).

3.2.2.4 Cotton-rayon material (tampon) method

Water-holding capacity was measured by a modified version of the cotton-rayon method of Walukonis et al. (2002). In this method an absorbent, approximately 3 g (Tampax superabsorbancy tampon, Procter&Gambler Co, Ohio, USA) was covered by a slice of pork sample and kept at 4 °C in a covered container for 3 hours. Then the

absorbent was weighted. WHC was calculated as the percentage of gained weight of the absorbent material (Walukonis et al., 2002).

3.3.3 Colour

Minolta (CM-3500d, Minolta Co. Ltd, Osaka, Japan) was used for colour measurements: *L*, a*, b** are taken after one hour blooming. Two readings were recorded from both sides of each sample. The average values were used for colour indicators of the samples. For enhancing the fraction of redness relative to *b** and *L** values, E values are calculated by using the following Equation 1 (Liu et al., 2003).

$$E = a^* / b^* + a^* / L^* \quad (1)$$

3.3.4 Data analysis

Drip loss and CIE colour values were analyzed by examination of the variance using the general linear model (GLM) of SAS (SAS Institute Inc., Cary, NC). The Duncan's test was used to test for differences among experimental groups.

Discriminant analysis was used for classification of quality groups. Discriminant Analysis (DA) is a multivariate statistical technique and commonly used to build a descriptive model of group discrimination based on observed predictor variables. The common objectives of DA are to investigate differences between groups, to discriminate groups effectively, to identify important discriminating variables, to perform hypothesis testing on the differences between the expected groupings and to classify new observations into pre-existing groups (Fernandez, 2002). In order to reduce the dimensions of data and make the discrimination models more robust, a variable selection procedure was conducted prior to the discriminant analysis. For this purpose, Stepdisc provided in the SAS software (Version 8, 1999, SAS Institute Inc., Cary, NC) was performed. The STEPDISC procedure performs a stepwise discriminant analysis to select a subset of the quantitative variables for use in discriminating among the classes. The significance level of a variable entering and staying in the model was chosen as 0.05.

Simple correlation presented in this study was calculated with the procedure of Pearson correlation coefficient of SAS.

3.4 RESULTS AND DISCUSSION

3.4.1 Water holding capacity measurements

Overall mean values of each WHC measuring methods of 4 pork quality classes is given in the following Figure 3.1. As seen from the figure, PSE meat had the highest drip loss of all 4 quality classes regardless of which measurement method used. At the same time, the normal quality meat, RFN, had almost the smallest drip loss values, which was an expected result. The second best WHC value is for PFN, which has the same textural property as RFN. The third best is RSE meat, which is a moderate form of PSE meat class. Our results were similar to Lee et al. (2000). In their study, PSE meat had the highest % Drip Loss (PD) and the RFN had lower PD (% drip loss) than other groups excepting DFD (Dry, Firm, Dry) meat. Our samples did not include DFD meat. It was reported by Hambrecht (2004) that DFD meat was frequent in cattle although it could occur in pigs, too. The results showed large variation in the WHC of samples, especially for the cotton-rayon and filter paper methods which had the highest standard deviation compared to the other two (bag and centrifuge) methods. In the study of Bertram et al. (2002), a large variation in WHC was reported among the meat samples used in the study, varying from 0.56 to 15.33% with an average drip loss of 5.88 %, as determined by the Honikel bag method. Also in the study of Otto et al. (2004), all measurement of drip loss showed a high coefficient of variance. It should be kept in mind that *Longissimus dorsi muscle* is a long muscle and could show different quality properties along the muscle at the same time. It implies that variations could be seen along the same muscle (Lundstrom and Malmfors, 1985). The filter paper method suffers from a large dependence on the roughness and composition of the meat surface where the filter paper is applied. This is also reflected in a very large standard deviation (Brondum et al., 2000).

For the first two methods, the results are more correlated because they are a continuation of each other. The centrifuge method showed a similar trend between the different classes. The filter paper and cotton-rayon methods have highest mean values compared to the other two methods. Actually having higher mean values between groups is good for classification. It gives noticeable differences among the groups. When

compared to bag method and centrifugation methods, both of the methods (filter paper and cotton-rayon) are rapid and time saving methods. The results showed that there were different mean values between the classes.

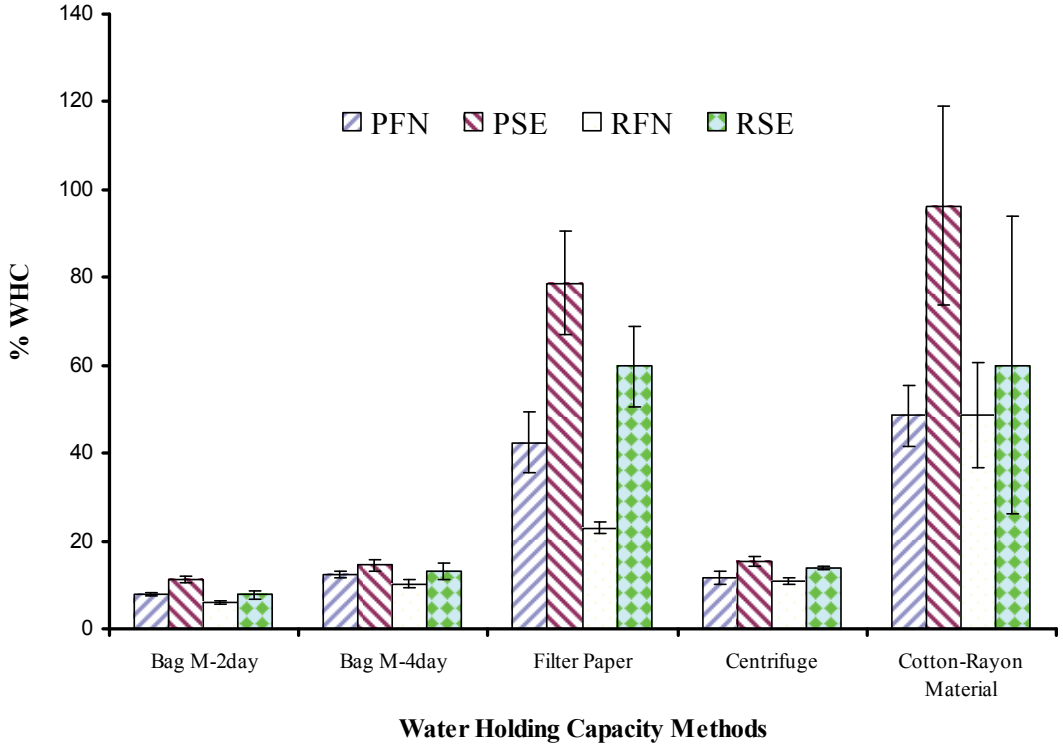

Figure 3.1 Overall mean-values of each WHC measurement values of 4 pork quality classes.

Correlations between each of the WHC measurement methods are shown in Table 3.1. There was a high correlation between the Bag M-2 day and the Bag M-4 day methods. These two methods provide similar descriptions for the WHC of meat, and therefore, may be used interchangeably (Otto et al., 2004). However, the correlations between the other WHC measurement methods were not consistent and generally low. These relatively low correlations were because the different methods represent different information about the WHC property of the meat samples (Brondum et al., 2000). While the drip loss reveals the amount of free water that exudes under the force of gravity from

the muscle fibres, the filter paper method measures the water that is extracted by applying a capillary force to the meat. The centrifuge method subjects meat samples to centrifugal force involving high speeds and gravitational forces. This method measures free water plus some other bound water exudate that is extracted by centrifugal forces. So, it is expected to have more WHC % than bag method. However it was not the case. It was almost parallel results as those of bag methods. Both the bag and centrifugation methods are in the same group named as gravimetric methods. However the centrifugation method is expensive and needs special equipments and time (Kauffman et al., 1986). In the study of Kristensen and Purslow (2001), it was reported that no changes in total water content of the meat were observed which could explain changes in WHC during ageing. These observations are consistent with the hypothesis that degradation of the cytoskeleton slowly removes the linkage between lateral shrinkage of myofibrils and shrinkage of entire muscle fibres, so removing the force that causes flow into the extracellular space. So, increasing WHC could be observed in later periods of storage. So, gravimetric (bag and centrifuge methods) results were close to each other.

In the cotton-rayon method, absorbed water is measured for prediction of drip loss (Walukonis et al. 2002). It was noticed that the correlation coefficient between WHC cotton-rayon material and filter paper methods had same values for Pale (PFN and PSE) and Red (RFN and RSE) pork meat groups namely as 0.48 and 0.61, respectively. This could be a good indication for cotton-rayon material and filter paper methods providing similar information to classify meat samples into two groups. Also both of the methods belong to the same group of WHC measuring methods which is absorbent methods (more detailed information is given in section 2.5.3.2 Absorbent methods). Both of the methods seem to be better methods for measuring WHC especially for grouping into two groups.

The correlation between colour values and WHC measurement results was studied (not presented here). Since it was concluded and agreed with van Laack et al. (1994) and Warris and Brown (1987) that pork colour and WHC are not closely related. Although colour and WHC appear to be associated, their specific biochemical properties seem to vary independently. So, there is growing evidence that colour is not a reliable predictor

of firmness and exudativeness (Swatland 1987; Kauffman et al., 1992; van Laack et al., 1994).

Table 3.1 Correlation coefficients between each WHC measuring method for each pork quality class.

Meat Classes	WHC measuring methods	Bag M. 2 day	Bag M. 4 day	Centrifuge	Cotton-rayon material	Filter Paper
PFN	Bag M.2 day	1.00				
	Bag M.4 day	**0.93**	1.00			
	Centrifuge	0.19	0.17	1.00		
	Cotton-rayon	0.15	0.07	0.30	1.00	
	Filter Paper	0.40	0.34	0.16	**0.48**	1.00
PSE	Bag M.2 day	1.00				
	Bag M.4 day	**0.97**	1.00			
	Centrifuge	0.14	0.27	1.00		
	Cotton-rayon	0.03	0.34	-0.13	1.00	
	Filter Paper	0.58	0.54	0.38	**0.48**	1.00
RFN	Bag M.2 day	1.00				
	Bag M.4 day	**0.91**	1.00			
	Centrifuge	0.34	0.47	1.00		
	Cotton-rayon	0.52	0.44	0.53	1.00	
	Filter Paper	-0.18	0.06	0.05	**0.61**	1.00
RSE	Bag M.2 day	1.00				
	Bag M.4 day	**0.70**	1.00			
	Centrifuge	0.41	0.36	1.00		
	Cotton-rayon	0.38	0.33	0.26	1.00	
	Filter Paper	0.30	0.07	0.23	**0.61**	1.00

3.4.2 GLM Duncan's test for each WHC measurement method

The GLM Duncan's test was performed for each parameter to see if the contrast between the different groups was significant. The results are shown in Table 3.2. WHC results indicate some significant differences between two or three groups, but none of them can separate alone the four classes successfully.

Table 3.2 GLM Duncan's test results for each WHC.

WHC measuring methods	Meat Classes			
	PFN	**PSE**	**RFN**	**RSE**
Bag Method- 2day	7.77 b*	9.62 a	7.58 b	7.15 b
Bag Method- 4day	9.85 b	11.12 a	9.67 b	9.08 b
Centrifuge	13.18 b, a	14.27 a	12.64 b	13.18 b, a
Cotton-rayon Material	68.27 c, b	113.32 a	55.98 c	78.16 b
Filter paper	30.38 b	48.75 a	20.11c	35.23 b

*** The means with the same letters in each row are not significant different at 0.05 significant level.**

From all 5 methods, only filter paper and cotton-rayon material methods had 3 different groupings. Filter paper can differentiate RFN and PSE separately but PFN and RSE were grouped as the same group. Cotton-rayon material method can differentiate PSE and RFN separately but RFN can be grouped also in PFN and RSE can be grouped in PFN. So, a little better classification was done by filter paper when compared to cotton-rayon material method.

3.4.4 Discriminant analysis of WHC measurement results

Although the ideal discrimination model should be able to separate the four quality groups with acceptable accuracy, it may also be more practical to classify the meat samples into P (PFN and PSE) and R (RFN and RSE) or FN (PFN and RFN) and SE (PSE and RSE) groups. Since according to GLM results, it is not possible to have 4 different classifications, Discriminant procedure was performed for only two group classification purposes to select the suitable WHC methods. The result of these two grouping was shown in the Table 3.3a.

Generally PSE could represent the worst pork meat quality whereas RFN could stand for the best pork meat quality. Consequently, it was studied to group four quality classes into different two groups such as PSE vs. (PFN, RFN, RSE) and RFN vs. (PSE, PFN, RSE) groupings. The result of these two groupings was shown in Table 3.3b.

From Table 3.3a, the results showed that 61% accuracy for classification into P-R grouping by the 2-day method and cotton-rayon material methods, and 64% accuracy for classification into FN-SE grouping by cotton-rayon material method were achieved. It could be said that cotton-rayon material was good enough for both kinds of classification (P-R and FN-SE). The study of Otto et al. (2004) reported that although drip loss has been measured for years, an international standard procedure is not available. As explained by Kauffman et al. (1993), the presence of RSE pork is the reason why the methodology does not work. Indeed, since the colour of RSE is similar to RFN pork, most chemical or physical procedures have failed to differentiate RSE from RFN. We had better results when trying to classify into 2 groups instead of 4. Nevertheless, for our experiments we used the 4 quality classifications as identified by the expert at the cutting house and there were discrepancies noted within each classification. The variation in the samples was high. For example, it was noticed that not all PSE samples had the same quality attributes, and in some cases, not all classified meat exhibited the expected quality attributes. It was mentioned by Hambrecht (2004) that quality classes such as PFN, PFE, and PFD have been described in literature but these are of less importance because of their relative low frequency. This could suggest that there must be more work done to study on better classification visually and instrumentally.

Cotton-rayon material or tampon method was introduced by Walukonis et al. (2002) for predicting WHC in early postmortem muscle. When all the WHC measuring methods are grouped according to the force applied, filter paper and cotton-rayon material go in the same group as capillary action or absorbent methods (as reviewed in "2.5.3.2 Absorbent methods"). From all WHC measuring methods, it can be said that only filter paper and cotton-rayon material methods had higher potential discriminatory power. Both methods could be used to develop discrimination models. The similar results can be seen from Table 3.3b. Cotton-rayon material method and filter paper method were good to classify pork meat samples into PSE vs. others and RFN vs. others by 70% and 65%, respectively.

Table 3.3a Discriminant procedure for all WHC measuring methods for classification into P–R and FN-SE groupings.

WHC measuring methods	*% Overall-cross validation accuracy*	
	P – R grouping	***FN-SE grouping***
Bag Method- 2day	61	**56**
Bag Method- 4day	**57**	**48**
Centrifuge	**57**	**55**
Cotton-rayon Material	61	64
Filter paper	**58**	**61**

Table 3.3b Discriminant procedure for all WHC measuring methods for classification into PSE vs. others and RFN vs. others groupings.

WHC measuring methods	*% Overall-cross validation accuracy*	
	PSE vs. others grouping	***RFN vs. others grouping***
Bag Method- 2day	64	56
Bag Method- 4day	59	49
Centrifuge	61	56
Cotton-rayon Material	**70**	63
Filter paper	61	**65**

3.4.4 Colour measurements

Figure 3.2 shows the 4 quality classes and their mean values of *L*, a*, b*,* and E values. PSE meat had the highest *L** value as expected. At the same time, the normal quality meat, RFN, had the smallest *L** value which was also expected.

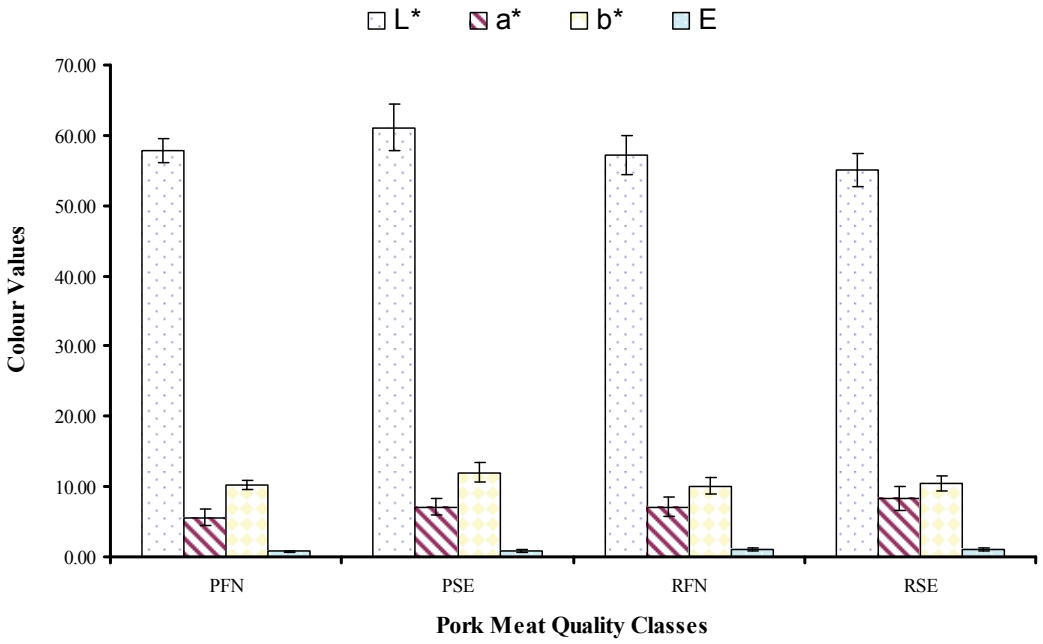

Figure 3.2 Overall *L, *a**, *b** and E values of 4 pork quality classes.**

3.4.5 GLM Duncan's test for each colour values

The GLM Duncan's test was performed for each parameter to see if the contrast between the different groups was significant. The results are shown in the following Table 3. 4. The four groups of meat do have significant differences between two or three groups according to all colour values, but none of them alone can separate the four classes successfully. To illustrate *L** value can differentiate PSE, RSE meat separately and PFN + RFN meat in the same group. For *a** value, there is 3 classes that are PFN, PSE+RFN, and RSE. For *b** value, there are two groups that are PSE, and PFN+RFN+RSE. For E value, there are two groups that are PFN+PSE and RFN+RSE.

Another point to mention here is that for *L** values between 40 and 60 are common for fresh pork. The greater the value (L*), the lighter the sample (Ken and Fedler, 2004). In our study, *L** values for 3 classes of pork samples that were PFN, RFN and RSE were between 40 and 60, except PSE meat that was over 60.

Table 3.4 GLM Duncan's test results for each colour values.

Colour measurements	Meat Classes			
	PFN	PSE	RFN	RSE
L* **value**	57.85 [b]	61. 11 [a]	57.16 [b]	55.02 [c]
a* **value**	5.63 [c]	7.13 [b]	7.10 [b]	8.30 [a]
b* **value**	10.23 [b]	12.05 [a]	10.14 [b]	10.43 [b]
***E* value**	0.79 [b]	0.82 [b]	1.08 [a]	1.08 [a]

*** The means with the same letters are not significant different in each row at 0.05 significant level.**

3.4.6 Classification according to colour results

Discriminant procedure was performed to examine the results when meat samples classified according to colour attributes are grouped together. As shown in the Table 3.5, totally 66% accuracy was gained for classification into FN-SE grouping (70% FN and 62% SE). For FN-SE grouping chosen parameters were *a** and E values. For grouping into Pale-Red class, totally 83 % accuracy was obtained (86% Pale +78% Red). We had highest result for grouping according to colour differentiation (pale and red) because we had colour values such as *L*, a** and *b** values. Of course, those results were not taken as based on only one of the parameters. Apparently, it is not easy to separate the four groups of meat using any single chromatic variable (Xing et al., 2007), or other quality parameters alone.

Table 3.5 Stepdisc and discriminant procedure for all WHC measuring methods for classification into P-R, and FN-SE groups.

	P – R grouping	FN-SE grouping
Overall-Cross validation accuracy	83 %	66 %
Chosen parameters	*L*, a** values	*a** and E values

3.5 CONCLUSIONS

The present study aimed to compare some of the drip loss measurement methods (bag method, centrifuge, cotton-rayon material and filter paper) and to investigate the relationships among CIE colour values for the classification of pork meat samples into 2 groups such as P-R or FN-SE groups. Filter paper and tampon methods may be possible to separate the exudative (SE) meat from non-exudative (FN) meat. From all quality groups (PSE, PFN, RFN, and RSE), PSE meat had highest WHC from the rest quality groups according to any WHC method. From CIE values, *a** and E were chosen for FN-SE grouping and for P-R groupings *L*,* and *a** were chosen.

The results showed that 83% accuracy for classifying the pork samples into 2 (P-R) groups and 66% for classifying into FN-SN groups based on colour parameters were achieved. The accuracy for classification into FN-SE groups according to WHC measuring methods was 64% by cotton-rayon material and 61% by filter paper method. With further studies use of cotton-rayon material or filter paper with a combination of other quality attributes could be improved for classification into 4 defined quality classes.

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CONNECTING TEXT

Almost all pork meat producers and processors agree on the fact that the main quality problems facing the pork meat industry today are insufficient water holding capacity and poor colour. Both of those parameters affect consumer's decision to purchase the meat product or not. There has been an increasing focus on drip loss as a quality parameter in meat products over the years. This interest has created several methods for determination of this quality parameter. Consequently, in the previous chapter, some of those water holding capacity measuring methods were studied for comparison purposes.

The next step was to investigate visible characteristic of pork meat, in other words, colour properties of different pork meat quality classes. In this study, the possibility of using visible spectroscopy (an objective and non-destructive system to assess the quality of fresh pork meat) to classify different quality classes of pork meat was investigated.

Some part of the results has been published in the Journal of Food Engineering in 2007: *Xing, J., M. Ngadi, A. Gunenc, S. Prasher and C. Gariepy, 2007. Use of visible spectroscopy for quality classification of intact pork meat, J.Food Eng., 82: 135-141.*

CHAPTER 4: USE OF VISIBLE SPECTROSCOPY FOR PORK QUALITY CLASSIFICATION

4.1 ABSTRACT

The objectives of this study were to study the potential of using visible spectroscopy to classify different quality classes of pork meat and to determine specific reflectance ratios to discriminate pork meat samples according to different quality classes. For this purpose, four different quality pork samples were studied; namely RFN (Red, Firm, Non-exudative), RSE (Red, Soft, Exudative), PFN (Pale, Firm, Non-exudative) and PSE (Pale, Soft, Exudative). The reflectance spectra of all pork samples were acquired with a Minolta CM 3500d Spectrophotometer in the range of 400 to 700 nm. The data analysis showed that it was possible to separate pale pork meat from red pork meat. In addition PFN meat was distinguishable from PSE pork meat. Also, the reflectance ratios of some wavelengths were suitable to classify pork samples into Pale and Red meat groups.

4.2 INTRODUCTION

Among the pork meat quality attributes, colour has special significance because it greatly affects consumers' decision when purchasing meat (Judge, 1989). Unfortunately, pork colour is difficult to measure because the colour of a meat cut, even within the same muscle, is commonly not uniform (AMSA, 1991). Normal quality pork is described as reddish-pink, firm and non-exudative, RFN, (red, firm, non-exudative). Different combinations of undesirable colour, texture and water holding capacity define different quality classes such as DFD (dark, firm, dry), PSE (pale, soft, exudative), RSE (red, soft, exudative) (Chan et al., 2002), and PFN (Pale, Firm, Non-exudative) meat (van Laack et al., 1994). It was reported by Cannon et al. (1996), during a Pork Chain Quality Audit survey in 1996, that packers registered a 10% incidence of PSE pork and a 4% incidence of DFD pork. The relationship between PSE and DFD quality defects and their respective colours influenced the industry to assign visual colour scores to carcasses.

Generally, fresh pork colour is visually evaluated by using either the Japanese Pork Colour Standards (JPCS) (Nakai et al., 1973) or National Pork Producers Council

Standards (NPPC, 1996) as a reference. Although those standards are useful, visual colour evaluation may change from one evaluator to another one and may be expensive (Tan et al., 2000). Therefore, the use of instrumental colour evaluation has gained much more attention because of its speed, consistency of measures, and potential use for on line sorting. For instance, CIE *L*, a** and *b** values (CIE, 1978) is an international standard for colour measurement. The 3-dimensioanl scale *L*, a** and *b** imitates the perception of colour by the human eye, and defines colour appearance in a way that can be readily understood (McGuire, 1992).

Spectroscopy has been studied extensively for determining properties of agricultural products because it is non-destructive, free of chemical preparation and has a fast inspection speed. When compared to plant materials, meat products have been studied less (Chan et al., 2002). Also, from all studies conducted to predict the quality of fresh meat, most attention has been focused on the prediction of beef tenderness, by using near infrared spectroscopy (Ru and Glatz, 2000; Hildrum et al., 1995; Liu et al., 2003; Shackelford et al., 2005). There are reports for predicting pork meat quality attributes such as drip loss and pH (Geesink et al., 2003; Liu et al., 2003; Forrest et al., 2000; Josell et al., 2000). There are not many studies reporting on classification of pork meat into different quality classes. Few studies are available in the prediction of colour characteristics from NIR measurement (Liu et al., 2003).

Reflectance measurement procedure is fast and relatively simple. Muscle structure, surface moisture, fat content and pigment concentration have an affect on the reflectance measurements (Hunt et al., 1991). More studies focused on estimation of deoxymyoglobin, oxymyoglobin and metmyoglobin. Hunt (1980) summarized equations of estimation of the myoglobin forms. However, many studies require only the detection of product colour differences rather than estimation of myoglobin forms.

Reflectance differences between wavelengths (630-580nm) or the ratio of 630/580 nm (Strange et al., 1974) have been useful in experiments where redness differences exist or decrease. Also, the American Meat Science Association (1991) indicated that the ratio of reflectance at 610 nm and 525 nm was an indicator of the percentage of myoglobin that was in the oxymyoglobin state.

A bright pink (red) colour is related to oxmyoglobin, while the colour of myoglobin is purple and metmyoglobin is more grayish or brownish pink (Lindahl et al., 2001). However, brown colours are difficult to measure instrumentally. Also, for meat products, it is often easier to measure a lack of redness or other normal colour. Lack of colour uniformity even within a slice of product, because of inherent muscle properties or processing techniques, causes colour measurement problems (Hunt and Kropf, 1985). Actually there is great number of factors affecting colour changes. Those factors make it difficult to define accurately and to follow meat colour changes.

Thus, the objectives of this study were to analyze how different reflectance ratios were useful to classify meat samples into different quality classes, and to investigate the potential of using visible spectroscopy for the classification of pork meat.

4.3 MATERIALS AND METHODS

4.3.1 Sampling and sample preparation

Pork loins were obtained from a local commercial cutting house, OLYMEL Plant in Quebec, Canada. A trained employee who had more than 10 years working experience selected the fresh pork samples from different quality groups namely PFN, PSE, RFN and RSE. The samples were shipped to the Food Engineering Laboratory at McGill University in Quebec. The samples were sliced into chops with a thickness of 1 cm and the experiments were conducted at room temperature (21±1 ºC).

A total set of 120 pork samples were used for Vis-spectral measurements. The samples were all obtained at 24 hr postmortem from the loin eye area.

4.3.2 Spectral data acquisition

The spectral reflectance of each pork sample was acquired with a Minolta CM 3500d spectrophotometer (Minolta Co. Ltd., Japan), in the wavelength region between 400 and 700 nm with 10 nm increment. A white tile (Minolta 13371004) was used as the white reference. For the dark measurement, the detector was covered by a black chamber provided by Minolta. Samples were placed in a sample cup equipped with a quartz

window with a diameter of 50 mm. In total, 6 readings were taken for each meat slice from both sides in order to get rid of the influence of the muscle fiber orientations on the spectrum and colour. The average of the 6 readings was recorded for each pork meat sample. Before taking the readings, the sliced meat samples were bloomed for 1 h (Honikel, 1998).

4.3.3 Data analysis

Discriminant Analysis (DA) was used to classify each observation into one of the 4 quality groups. In order to reduce the dimensions of data and make the discrimination models more robust, a variable selection procedure was conducted prior to the discriminant analysis. For this purpose, Stepdisc provided in the SAS software (Version 8, 1999, SAS Institute Inc., Cary, NC) was performed. The Stepdisc procedure performs a stepwise discriminant analysis to select a subset of the quantitative variables for use in discriminating among the classes. The significance level was set at 0.05.

4.4 RESULTS AND DISCUSSION

4.4.1 General view of the reflectance spectra

The average spectral reflectance for each quality class (PFN, PSE, RFN and RSE pork meat) is shown in Fig.4.1. The pale meat, PSE group, has a higher reflectance value than the red meat because of its brightness. As shown in the Fig.4.1, PSE pork consistently has a higher spectral reflectance than the other three meat quality groups, and the mean spectral reflectance of RSE group is consistently lowest. RFN and PFN groups are almost on top of each other. RFN and PFN are very closely matched up to about 630 nm at which point they start to separate from each other. From this observation, it may be concluded that firm and non-exudative pork meat (FN group; PFN and RFN) have similar spectral reflectance in the visible range although their colour is different.

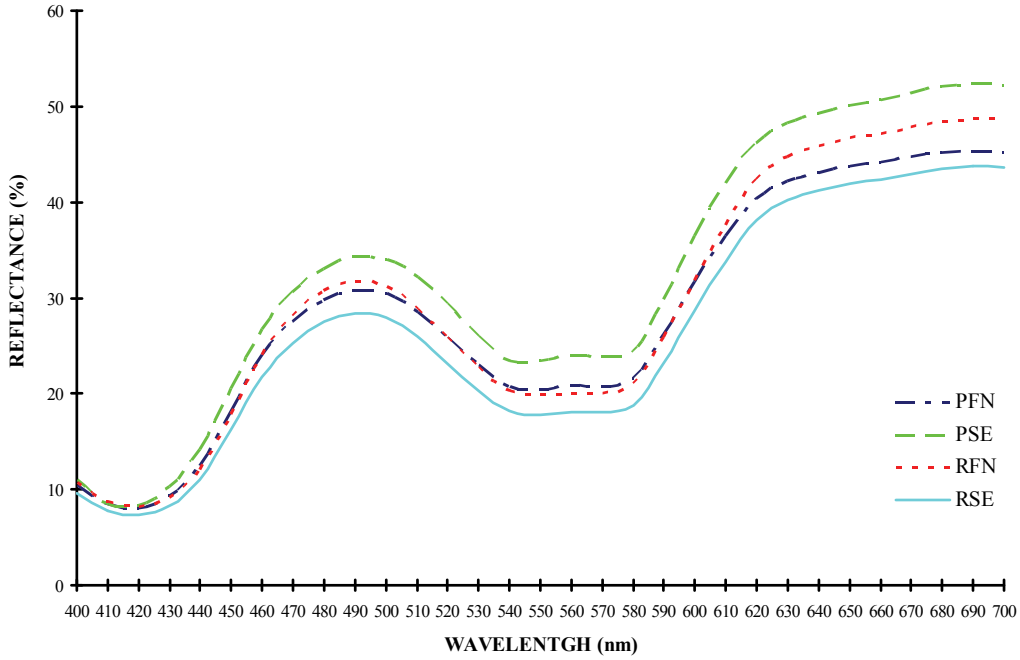

Figure 4.1 Mean reflectance of each class of 4 pork meat groups.

However, for soft and exudative pork (SE group; PSE and RSE) despite the fact that both PSE and RSE are soft and exudative, they give different reflectance values at visible spectral range and they have different colour characteristics (one is pale, and the other is red). It could be explained by the effect of light scattering because light scattering increases with free water, and tissue appears more lighter (Brewer et al., 2001). Consequently as shown in the Figure 4.1, the colour difference is more obvious for SE meat (PSE, and RSE) but not for the FN group (PFN and RFN).

The Duncan's test (in GLM procedure) revealed that classification into 4 quality classes (PFN, PSE, RFN, and RSE) was not possible at the visible range except in the range of 620-630 nm. In the range of 620-630 nm, we have significant differences in the 4 quality classes. The other ranges can be used for the classification into at least 2 or 3 groups. PSE and RSE groups can be separated more easily than RFN and PFN groups (in the range of 430-610 nm). Reflectance ranges except 620-630 nm can be used for the

classification into at least 2 or three groups. One might conclude that the mean spectra of FN groups are almost on top of each other and SE groups are always far from each other. This suggests that pork meat samples will be more easily classified into FN-SE or pale–red classes rather than into 4 classes.

There is an interesting spectral feature observed at around 560 nm from the Figure 4.1. The little bump at around 560 nm was seen in both of PSE and PFN mean spectra. Both of them are pale groups. It is believed that the reflectance at 560 nm is to be caused by myoglobin (Brondum et al., 2000; Liu and Chen, 2001). This result might imply that the content of myoglobin might be higher in Pale groups than Red groups, but it was not possible to successfully separate the meat into the different quality groups based on this feature alone.

4.4.2 Discriminant analysis

Although the ideal discrimination model should be able to separate the four qualities with an acceptable accuracy, it may be more practical to classify pork meat samples into P (PSE and PFN) and R (RFN and RSE) or SE (PSE and RSE) and FN (PFN and RFN) groups. Discriminant procedure was performed for those purposes, and stepdisc was used to select the suitable wavelengths. As shown in the Table 4.1, we had 67% accuracy for classification into 4 groups, 85% accuracy for classification into pale-red, and 61% accuracy for classification into FN-SE groups. We had the highest accuracy in the classification into pale-red groups with a total error of about 15 %.

Table 4.1 Discriminant procedures for classification into 4, P-R, and FN-SE groups.

	Classify into		
	4 Groups	***P–R Groups***	***FN- SE Groups***
Cross validation accuracy	67 %	85 %	61 %
Chosen wavelengths(nm)	420, 580, 450, 400 and 490	500, 430, 550, 570, and 510	410 and 420

4.4.2.1 Discriminating the pale and red classes

The classification results of pale and red classes using the wavelength 500, 430, 550, 570 and 510 nm are given in the following Table 4.2

Table 4.2 Classification of pork meat samples into P and R classes using five wavelengths (500, 430, 550, 570 and 510 nm).

From	**Classify into**		**Total**
	P (PFN+PSE)	***R (RFN+RSE)***	
P	49 ***81.67***	11 18.33	60 100.00
R	8 13.33	52 ***86.67***	60 100.00
Total	57 47.50	63 52.50	120 100.00

It can be said that possible misclassified samples are likely from FN (PFN and RFN) group than from the SE (RSE and PSE) group because RFN and PFN mean reflectance spectra were almost together (as shown in the Fig.4.1). This may be explained by the effect of light scattering. Some factors like muscle structure and surface moisture can cause light scattering. Also, packing of each meat sample into the sample cup may cause deformation of muscle fiber orientation effects and change the penetration of the light through the meat sample (Cozzolino et al., 2003). Generally FN pork meat is firmer and contains less moisture than SE pork meat. Therefore, there was less light scattering in the FN tissues than in the SE tissues. This might explain why there is less misclassification in SE than in FN group. Also in Fig.4.1, PSE and RSE meat always had mean reflectance values far from each other.

4.4.2.2 Discriminating samples within the pale class

The reflectance spectra data suggests that the PFN class of meat may be differentiated from the PSE quality of meat. For this purpose, Stepdisc was performed for the variable selection for the discriminating analysis. The selected wavelengths were 630, 430, 450, 520 nm. Table 4.3 gives the classification results using the reflectance at the selected wavelengths. About 93.33% of the PFN samples were correctly classified. Slightly less classification accuracy (73.33%) was obtained for the PSE pork meat.

Table 4.3 The discriminant procedure; classification summary within the P groups (PFN and PSE) with using chosen wavelengths at 630, 430, 459 and 520 nm.

From	Classify into		Total
	PFN	***PSE***	
PFN	28 93.33	2 6.67	30 100.00
PSE	8 26.67	22 73.33	30 100.00
Total	57 47.50	63 52.50	120 100.00

4.4.2.3 Discriminating samples with using some reflectance ratios

Reflectance measurements are affected by muscle structure, surface moisture, fat content and pigment concentrations. Many of these effects may be corrected by using ratios of reflectance at different wavelengths or by using differences between reflectance at different wavelengths. The reflectance ratio of 630/580 (Strange et al., 1974) has been useful in experiments where redness differences exist, or decline. By using GLM, we had significant differences in classification of 4 meat classes into two groups as red and pale classes. The GLM result is given in Table 4.4.

Table 4.4 The GLM results of reflectance ratio of 630/580 for classification all pork samples.

Meat Class	Mean value	N
PFN	1.96 [b*]	30
PSE	1.99 [b]	30
RFN	2.13 [a]	30
RSE	2.16 [a]	30

***The same letters are not significant different**

Then discriminant procedure and stepdisc were employed for other ratios of all Vis-ranges. The chosen ratios were 500/560, 620/580 and 620/400 by stepdisc. We had 83% accuracy in the classification of pork samples according to pale and red groups. The results are given in Table 4.5.

Table 4.5 The classification into pale and red groups according to chosen reflectance ratios of 500/560, 620/580 and 620/400.

From	Classify into		Total
	P (PFN+PSE)	***R (RFN+RSE)***	
P	***48***	12	60
	80.00	20.00	100.00
R	9	***51***	60
	15.00	***85.00***	100.00
Total	57	63	120
	47.50	52.50	100.00

4.5 CONCLUSION

The aim of this study was to investigate visible spectroscopy for classification of different pork meat quality classes and to examine any suitable specific wavelengths or reflectance ratios to discriminate the pork meat samples. The results showed that it is possible to separate the P (pale) classes of pork meat samples from the R (red) classes with an accuracy of about 85%. It is also possible to separate P meat from R meat just using specific reflectance ratios with an accuracy of about 83%. However, only visible spectral information is not sufficient to separate RSE from RFN pork meat.

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CHAPTER 5: CONCLUSIONS AND RECOMMENDATIONS

5.1 GENERAL CONCLUSIONS

In this study, some of pork meat quality attributes for the classification of pre-determined pork classes were studied. Mainly, exudativeness and colour of pork meat were investigated. Also potential of using visible spectral reflectance values for discriminating the pork groups was included. Based on the experimental data and related analysis and discussion, we can make the following interpretations and draw the following conclusions;

I. PSE pork meat class had always highest drip loss value from all groups PFN, RFN and RSE groups.
II. From all studied WHC measuring methods, cotton-rayon material and filter paper methods were good to separate the exudative meat from non-exudative meat.
III. Based on colour values (L^* and a^*), we had 83% accuracy for the classification of pork meat samples into P-R groups.
IV. In the study of visible spectroscopy for classification of pork meat samples, we had 85% accuracy in separating the pork samples into P and R groups. According to the discriminating analysis, certain wavelengths were chosen such as 500, 430, 550, 570, and 510 nm.
V. Also some wavelength ratios were found useful for classification. They were 500/560, 620/580 and 620/400.
VI. All experimental results showed that we had succeded in grouping the pork samples into two groups based on colour or exudativenes, in another words, P-R or FN-SN groups. But the results were not good to separate them into 4 defined quality classes.

5.2 RECOMMENDATIONS

Further research is recommended to focus on the following points;

For WHC measuring methods, further study should be done on absorptive methods such as cotton-rayon material and filter paper methods. Both of them do not require much investment and time and they are easy to apply.

Finding an international standard for classification of pork meat is very crucial. Also, before reaching this goal, more work should be done on pork meat quality definition for detailed parts of pork carcasses because there are many factors defining total quality.

To reach ideal discrimination of pork meat classes according to quality, there must be a method that might combine more than one attributes. More studies should be done on combining of different methods; could be including sensory analysis and technological analysis together. Even every genotypes of pig should be investigated thoroughly to define each specific character of pig and then potential effects on the pork meat quality.

APPENDIX

1. ANOVA table for measuring WHC by Bag method 2 day

Source	DF	Sum of squares	Mean squares	F	Pr>F
Model	23	600.761341	26.120058	7.05	<.0001
Error	156	577.671390	3.703022		
Corrected Total	179	1178.432731			

2. ANOVA table for measuring WHC by Bag method 4 day

Source	DF	Sum of squares	Mean squares	F	Pr>F
Model	23	1205.300593	52.404374	12.32	<.0001
Error	156	663.386840	4.252480		
Corrected Total	179	1868.687433			

3. ANOVA table for measuring WHC by Centrifuge method

Source	DF	Sum of squares	Mean squares	F	Pr>F
Model	23	495.909589	21.561286	3.20	<.0001
Error	156	1050.191910	6.731999		
Corrected Total	179	1546.101499			

4. ANOVA table for measuring WHC by Tampon method

Source	DF	Sum of squares	Mean squares	F	Pr>F
Model	23	207817.3865	9035.5385	6.32	<.0001
Error	156	222885.9617	1428.7562		
Corrected Total	179	430703.3482			

5. ANOVA table for measuring WHC by filter paper

Source	DF	Sum of squares	Mean squares	F	Pr>F
Model	23	103727.7071	4509.9003	22.10	<.0001
Error	156	31831.0042	204.0449		
Corrected Total	179	135558.7113			

6. ANOVA table for *L*, a and *b** values**

Source	DF	Sum of squares	Mean squares	F	Pr>F
Model	11	1497.281469	136.116497	18.19	<.0001
Error	108	808.008390	7.481559		
Corrected Total	119	2305.289859			