COMPUTER VISION SYSTEM AS A TOOL TO ESTIMATE PORK MARBLING

A Dissertation
Submitted to the Graduate Faculty
of the
North Dakota State University
of Agriculture and Applied Science

By

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In Partial Fulfillment of the Requirements
for the Degree of
DOCTOR OF PHILOSOPHY

Major Department:
Animal Sciences

June 2017

Fargo, North Dakota

North Dakota State University
Graduate School

Title

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The Supervisory Committee certifies that this ***disquisition*** complies with North Dakota State University's regulations and meets the accepted standards for the degree of

DOCTOR OF PHILOSOPHY

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ABSTRACT

Currently pork marbling is assessed subjectively in the industry, because of the limited methods and tools that are suitable for the industry. In this dissertation, we are devoted to develop a computer vision system for objective measurement of pork which suits the industrial needs. Experiment 1 examined the possibility of using computer vision system (CVS) to predict marbling in a lab-based experiment using pork samples that were already trimmed of subcutaneous fat and connective tissue. Experiment 2 an industrial scale CVS was built to predict the 3^{rd} and 10^{th} rib pork chop's marbling. Experiment 3 the industrial scale CVS was tested in the meat plant and images of whole boneless pork loin were collected. The CVS predicted marbling were compared with subjective marbling score using crude fat percentage (CF%) as standard.

In experiment 1 subjective marbling score had a correlation of 0.81 with CF% while CVS had a 0.66 correlation. CVS has shown an accuracy of 63% for stepwise regression model and 75% for support vector machine model. These results indicate that CVS has the potential to be used as an tool to predict pork intramuscular fat (IMF)%. In experiment 2 the accuracy of CVS predicting pork chop CF% was 68.6% and subjective marbling was 70.1%. A drop of accuracy in predicting anterior chop CF% for both CVS and objective marbling score was observed when compared to posterior chop, this suggest that there is a discrepancy in accuracy between the anatomy location of samples collected. In experiment 3 the accuracy of CVS predicting boneless whole loin was 58.6% and subjective marbling score was 53.3%. In this research, CVS has demonstrated a consistency of accuracies using different pork samples. CVS has shown higher accuracy when predicting whole boneless loin IMF% when compared to subjective assessment.

ACKNOWLEDGMENTS

I would like to extend my sincere gratitude to all who helped make the completion of this dissertation possible. I would like to thank Dr. David Newman for giving me the opportunity to study abroad and obtain my doctoral degree in Meat Science and Muscle Biology. Through the four years of his advisory, he showed me true leadership, encouragement, and patience as I worked to complete this goal. I would also like to thank Dr. Eric Berg, Dr. Xin Sun, Dr. Pawel Borowicz, and Dr. Harlene Hatterman-Valenti, my committee members, for their guidance and assistance in learning during my time at NDSU.

Thanks to all the faculty members that I have had opportunity to work with or taken a class from. I have learned so much from the passion and expertise of all the people I worked with. It was truly an inspiration to me and I shall always remember to follow your lead and pass on the passion.

A special thanks goes to the Newman lab group (Dr. Jennifer Young, Dr. Xin Sun, Laura Bachmeier, Stephanie Schaunaman, and Rosemarie Somers) for the times we have spent together and all the pork chops that we have collected data from, this will be an experience that will haunt me for life.

My family has been a huge support to me throughout my pursuit of my Ph.D. Being on the other side of the world has its ups and downs. I am glad to have a supporting family that I can share my full experience with, knowing they have full confidence in me regardless of the challenges that I encounter. Without their support and understanding, none of this would have been possible.

DEDICATION

I would like to dedicate this dissertation to my father, Dr. Deng Chen Liu.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

ATP ....Adenosine tri-phosphate

Ca^{2+} ....Calcium ion

CAST ....Calpastatin gene

CF% ....Crude fat percentage

CF1..Category of crude fat percentage of 0 – 1.99 %

CF2..Category of crude fat percentage of 2 – 2.99 %

CF3..Category of crude fat percentage of 3 – 3.99 %

CF4..Category of crude fat percentage of 4 – 4.99 %

CF5..Category of crude fat percentage of greater than or equal to 5 %

CIELAB ....L*, a*, b* color space
- L* ....Black = 0, white = 100
- a*...negative = green, positive = red
- b*...negative = blue, positive = yellow

CO_2..Carbon dioxide

CVS..Computer vision system

DFD...Dark, firm, and dry

GLCM ....Grey-level co-occurrence matrix

H_2O ....Water

HSI ....Hue, saturation, intensity

IIMF ....Image intramuscular fat percentage

IMF ....Intramuscular fat

IMFresid...Residual (crude fat percentage minus estimated intramuscular fat percentage from regression model)

IMF% .... Intramuscular fat percentage

LED .... Light-emitting diode

NDSU .... North Dakota State University

NPB .... National Pork Board

O_2 .... Oxygen

pH .... Power of hydrogen

PSE .... Pale, soft, and exudative

PSS .... Porcine Stress Syndrome

RBF .... Radial basis function

RFN .... Red, firm, and non-exudative

RGB .... Red, green, blue

RIMF% .... Estimated intramuscular fat percentage from regression model

RIMF1 .... Category of estimated intramuscular fat percentage from regression model of 0 – 1.99 %

RIMF2 .... Category of estimated intramuscular fat percentage from regression model of 2 – 2.99 %

RIMF3 .... Category of estimated intramuscular fat percentage from regression model of 3 – 3.99 %

RIMF4 .... Category of estimated intramuscular fat percentage from regression model of 4 – 4.99 %

RIMF5 .... Category of estimated intramuscular fat percentage from regression model of greater than or equal to 5 %

ROI .... Region of interest

RSE .... Red, soft, and exudative

SMS .... Subjective marbling score

SVM..Support vector machine

US ....United States

Vis/NIR...Visible light and near-infrared region spectroscopy

WHC ....Water holding capacity

WLD ....Wide line detector

μ ....Mean

σ ....Standard deviation

CHAPTER 1. INTRODUCTION AND LITERATURE REVIEW

Introduction

Pork is currently the most consumed protein source globally (15.8 kg/capita/yr), followed by poultry (13.6 kg/capita/yr), beef (9.6 kg/capita/yr), and sheep and goat meat (1.9 kg/capita/yr) (FAOSTAT, 2014). Meat purchasing decisions are influenced more by product appearance, such as color and marbling, than any other quality factor (Font-i-Furnols et al., 2012). In 2015, Newman et al. reported that the average for pork subjective color score is 2.85 ± 0.79 on a 6-point scale, with 3, 19, 45, 26, and 7 % of samples having a color score 1, 2, 3, 4, and 5 respectively. The average for subjective marbling score (an estimate of intramuscular fat percentage) is 2.30 ± 1.07, with a distribution of 9, 47, 31, 10, and 3 % for marbling scores 1, 2, 3, 4, and 5 or above, respectively. This shows that there is variation in pork quality in the retail market throughout the US.

Currently pork color and marbling scores are determined by trained evaluators in the plant, which is subjective and lowly repeatable. It is also influenced by the condition of the evaluator, such as sickness or fatigue, or different environments, such as lighting or angle of viewing. In a laboratory environment, color and marbling can be assessed more objectively using a colorimeter, which can express color using L*, a*, and b*, and ether extract to determine the percentage of intramuscular fat (IMF). However, in 2013, Girolami et al. reported when regenerating a color by using the L*, a*, and b* values recorded by colorimeter, the color did not correspond to the true color of meat. Additionally, ether extract is a labor intensive and time-consuming procedure, which requires an actual sample that could sabotage the integrity of and potentially de-value the product. This suggests that a new modern technology that is rapid, accurate, non-invasive, and highly repeatable could be beneficial for both research and industry.

The potential of using computer vision system (CVS) in the food industry has long been recognized (Timmermans, 1998). With recent advances in hardware and software, CVS has been allowed to become a technology even more cost effective, more consistent, more rapid, and more accurate than ever before. A CVS is a system, which is composed of three main elements: camera, lighting system, and image analysis software. A CVS allows for the capturing, processing, and analyzing of images, which enables the assessment of a desired target in an objective, non-destructive manner. This technology has been applied for numerous usages in the food industry such as classification of types of cereal grains (Paliwal et al., 2001), color grading for apples (Nakano et al., 1992), and detection of bruises on strawberries (Nagata et al., 2006). In the beef industry, CVS has been utilized to objectively measure features of beef quality such as marbling and yield percentage using the "beef cam". Research has shown the potential of CVS in predicting beef color (Larraín et al., 2008), fat color, (Chen et al., 2010), tenderness (Sun et al., 2012; ElMasry et al., 2012), and marbling (Chen et al., 2010). With CVS successfully applied in many different fields with different goals, it only seems reasonable to use this technology in the pork industry as well.

In this chapter, pork quality will be defined, how each pork quality trait effects the others will be discussed, and current methods of measuring pork quality will be addressed. Next CVS will be introduced and how it has been applied to the industry will be discussed.

Quality

What is quality? Pork quality is a combination of many factors and cannot be defined based on one variable alone because each individual factor can be influenced by one or more other factors (Ben, 2011). The answer may also differ depending on what part of the pork industry you participate in: quality may be as simple as a pig that grows fast on minimal feed if you are the hog producer or as complex as the combination of visual attractiveness and eating satisfaction if you are the consumer. Eating satisfaction is influenced by the color, texture, and marbling of pork and is usually assessed by evaluating flavor, tenderness, and juiciness. For pork processors and retailers, the definition of quality can vary depending on what they focus on. Since the most important factor to consumer's when they are purchasing pork is color, good color (reddish-pink) would be considered good quality. Additionally, pork with low drip loss values would be more favorable to processors as there would be less weight loss from slaughter to package, which results in more profit. Additionally, pork with high water holding capacity produces less purge after packaging, which is more favorable to consumers.

The National Pork Board (NPB) has established pork quality standards in which it describes three types of pork quality based on the pork's color, texture, and ability to bind water: red, firm, and non-exudative (RFN); pale, soft, and exudative (PSE); and dark, firm, and dry (DFD). Pork that is RFN has a reddish-pink color, firm texture, and minimal exudation on the cut surface or in the package, which is visually attractive to the consumer. Ideally, it would be best to provide consumers with RFN pork. The reddish-pink color indicates that the myoglobin has gone through minimal amount of denaturing; a firm texture indicates integrity of the structure of muscle; and being non-exudative suggests that the meat is capable of binding or retaining water, resulting in minimal cook loss and pork that is juicy. Pork that is PSE is considered poor in quality, which contributes a large loss to the pork industry. In 1996, Cannon et al. reported that 10.2 % of carcasses were PSE and, in 2003, Stetzer and McKeith reported approximately 15.5 % of the pork produced in US had characteristics of PSE pork. A rapid pH decline while the carcass is still at a high temperature results in higher than normal myoglobin denaturation. This results in pork that is pale in color and has poor water holding capacity. Once cooked, PSE pork loses even more water and results in tough, dry pork and a poor eating experience. There are many factors that contribute to pork becoming PSE such as genetics, preslaughter stress, and carcass chilling. Pork that is DFD is considered to have good pork quality. The DFD pork is a result of a slow pH decline, which results in a higher ultimate pH compared to RFN and PSE. The structural proteins are a lot less denatured, which allows the pork to bind with more free water, resulting in less water on the surface, which causes it to look darker. Kauffman (1993) stressed the importance in taking greater advantage of high pH meat to gain profit for the processor and greater satisfaction for the consumer. However, due to its high ultimate pH, DFD pork typically has a shorter shelf life due to bacterial growth when compared to RFN and PSE (Faucitano et al., 2010).

Muscle to Meat

The "muscle to meat" conversion is a process of physical and biochemical changes after an animal is slaughtered that can be impacted by many factors. This process is affected by genetics, pre-slaughter handling, nutrition, short- and long-term stresses, and post-slaughter handling of meat.

In the living animal, the main function of skeletal muscle is to support and contract. Both contraction and relaxation of muscle consumes energy such as adenosine tri-phosphate (ATP), which is provided and replenished by the mitochondria. Under aerobic environments, muscles maintain energy through aerobic glycolysis, which converts one glucose molecule to 36 ATP, 6 carbon dioxide (CO_2), 6 water (H_2O), and heat. When the existence of oxygen is insufficient, muscles produce energy through anaerobic glycolysis, which converts one glucose molecule to 2 ATP and 2 lactate. The lactate is then transported by the blood to liver where it is converted back into glucose.

Conversion from muscle to meat is an enviable procedure, which involves numerous biochemical pathways; it is traditionally viewed as an anaerobic process that is largely influenced by degradation of glycogen to lactate and hydrogen ions. At harvest, muscle must adapt to new physiological circumstances to maintain ATP and homeostasis. As the animal is being bled, delivery of oxygen (O_2) is eliminated, which hinders mitochondria from contributing to ATP production. It also induces the pathway of anaerobic glycolysis, which transforms glucose into lactate. Because it is deprived of blood, the lactate accumulates within the muscle and results in pH decline. The insufficient amount of ATP then causes the onset of "rigor mortis", which is when the muscle stays in its contracted state. Onset and completion of rigor mortis normally occurs within the first 24 h after slaughter.

During this anaerobic process, different metabolic processes and enzymatic activities are involved and many factors can influence the process. It is also the critical process that ultimately influences the quality of pork. The amount of lactic acid that can be accumulated may differ due to differences in muscle type, glycolytic potential, and genetics (Fernandez et al., 1994; Ryu et al., 2005; Salas et al., 2017). Glycolytic potential can also be influenced by stress prior to slaughter. In 2004, Hamilton reported long transportation increased muscle glycogen in two different muscle when compared with short transport. Later on, Scheffler et al. (2013) reported that while low glycolytic potential may limit glycolysis and are associated with ultimate pH, high glycolytic potential does not predict low ultimate pH in pork. Genetic differences such as breed also influence glycolytic potential as Monin et al. (1987) reported that when compared to Large Whites, Pietrains, and Landraces, Penshires had much higher glycolytic potential in the fast white muscle. It is also reported that swine calpastatin gene (CAST) DD genotype has a lower glycolytic potential when compared with CC and CD (Boruszewska et al., 2016). Swine with

Porcine Stress Syndrome (PSS) gene are also more susceptible to hyperthermia under stress conditions (Fisher et al., 200a). It has been established when PSS occurs, pigs respond with limb muscle rigidity, increased anaerobic metabolism, and increased body temperature due to the "fight, fright, or flight reaction" (Bjustrom et al., 1995). It is generally believed that PSS is caused by a mutation in gene coding for the calcium ion (Ca^{2+}) release channel because, when abnormality of Ca^{2+} release occurs, the uncontrollable increase of intracellular Ca^{2+} causes muscle rigidity, hypermetabolism, hyperthermia, and metabolic acidosis (Bjustrom et al., 1995). The rate of lactic acid accumulation can be effected by the temperature of carcasses, as enzymatic activity decreases with the temperature. It is reported that a reduction in glycolysis occurs when utilizing blast chilling (Milligan et al., 1998; Springer et al., 2003). It is believed that heavier pigs show a greater tendency to develop PSE, as heavier carcasses take longer to cool due to the larger volume to surface area ratio.

Color

Color may be the most important factor that influences the appearance and attractiveness of pork to consumers (Lu et al., 2000; Leon et al., 2006; Valous et al., 2009) as consumer use inadequate color as an indicator of spoilage. When buying pork, consumers prefer pork that is lean in appearance, consistent in color, with little amount of water on surface or in-package (Mabry & Bass, 1998). Typically, consumers prefer pork that is darker versus lighter in color (Brewer & McKeith, 1999; Norman et al., 2003). However, meat acceptability depends on cultural aspect. Preferences for color may also vary between culture, as consumers from Australia (light red, lean), Korea (marbled), Poland (lean), and Taiwan (dark red, lean) all have different preferences (Ngapo et al., 2013). Japan, as one of the most profitable pork export markets for the US, prefers moderately firm, well marbled muscle with a darker color (NPPC color score 3-5) (Cravens, 2000).

The color of pork is constituted mainly by myoglobin (Hedrick et al., 1989). Myoglobin, a heme protein, is responsible of combining O_2 and gives meat its red color. While the association of color and freshness of pork is weak, color serves as a good indicator of the quality of pork. Pork with darker color were perceived to be more tender, more juicy, and less dry than lighter pork by both trained panel and consumers (Norman et al., 2003).

In the pork industry, pork color is often graded by professional trained evaluators, which grade the pork using the NPB standard color cards from grade 1 (pale white) to 6 (dark red). From the NPB benchmarking project in 2015 has suggested that the mean subjective color score values were 2.85 ± 0.79 when observed within the retailers or supermarkets meat case. This indicates that not only there is a great deal of pork quality variation in the retail meat case, but also slightly pale.

For meat science research, pork color is often measured and quantified by using a colorimeter which often express the attributes as L*, a*, and b*, where L* measures lightness to darkness (100 = white; 0 = black), a* measures redness to greenness (positive = red; negative = green), b* measures yellowness to blueness (positive = yellow; negative = blue). There are two commercially available colorimeters; Hunterlab (Hunter Associates Laboratory Inc., Reston, VA) or Minolta Colorimeter (Minolta Company, Ramsey, NJ). Most researchers use Minolta colorimeter (60 %) over Hunter (31.6 %) colorimeter under D_{65} illumination, which more closely resembles daylight and resulted in higher correlations with visual color scores (Tapp III et al., 2011). It has been well established that using L* as an assessment for pork color has a high correlation with subjective color assessment score (Warriss and Brown, 1993). In 2001, Brewer et al. concluded that L* was not prejudiced by bloom time, indicating it is the best value to determine if a carcass is predisposed to becoming PSE or DFD pork. In 1994, Laack et al. also reported, while dark pork (L* < 52.0) always has acceptable water-holding capacity (WHC), pale pork (L* > 58.0) does not always have unacceptable water holding capacity. This suggests that when assessing pork quality, it is best not to rely only on color but also other factors such as pH or WHC. In 2015, Newman et al. reported that the mean color values of pork chops in retail stores across the nation is 55.56 ± 3.63 for L*, 16.60 ± 2.30 for a*, and 10.33 ± 1.53 for b*. An L* value near 55 or below (Minolta) is associated with an NPB color standard score of 3.0 as a reference (NPB, 2001) which agrees with the subjective color results.

When using a computer vision system color can be expressed in different color space such as HSI (hue, saturation, intensity), RGB (red, green, blue), and Computer estimated L*, a*, b*. While the L*, a*, b* generated by computer would differ between illumination, settings of camera, it is reported to have a high correlation with Minolta L*, a*, b* (Sun et al., 2016).

Marbling

Marbling can often be referred as IMF content and is generally accepted that the degree of marbling has a positive influence on the sensory qualities of pork (Brewer et al., 2001; Cannata et al., 2010; Wood et al., 2004;). More specifically research has suggested that with an increasing of marbling in pork loin result in higher sensory score such as tenderness and juiciness of pork. Not only marbling has influence on the eating palatability, it is also a factor which influence consumers willing of buying as the amount of marbling, however, the preference of consumers toward marbling may differ between or within country. In the US, marbling has been reported to negatively affect consumer preferences and the acceptability of red meat (Brewer et al., 2001; Moeller et al., 2010) because chops with a greater degree of marbling appear to have more fat and lighter in lean color. However, the acceptability of marbling is more favorable and desired in Asiatic countries (Ngapo et al., 2007). Regardless of consumer's preference of marbling when considered as a visual factor, even when consumers are divided into "lean loin lovers" and "marbled loin lovers", both groups show a preference for highly marbled pork when tasting the cooked product where they cannot see the amount of marbling present (Furnols et al., 2012). In 2008, Meisinger suggested an industry target for IMF of 2 to 4 %, with the minimum level reflecting minimum eating satisfaction requirements and the maximum level reflecting the health concerns associated with consuming excessive fat. In 2015, Newman reported that in the current market, the average of subjective marbling within the retail market is 2.54 with a distribution of 9, 47, 31, and 10 % for marbling grades 1, 2, 3, and 4, respectively.

Current methods to measure IMF in pork are limited. In laboratory-based research, IMF can be determined by using crude fat extraction (AOAC, 1990); however, this extraction method is not only labor-intensively and time-consuming but requires using the actual samples making it impractical for the pork industry. The most common way to determine IMF in the pork industry is by subjective assessment. Assigning subjective marbling scores requires trained individuals assessing the pork loins using pork marbling standard cards (NPB, 2011) on a scale of 1 to 10 (1=devoid, 10=abundant). However, since this is a subjective measurement, variation can occur due to different evaluators, different lean colors, different genetics may cause different infiltration of IMF and distribution, and different environmental factors in the slaughter plant such as lighting.

pH

The pH of pork plays an important role and has a direct impact on pork quality traits such as WHC and color. In live animals, physiological pH of muscle is near 7.2 but, in the "muscle to meat" conversion, the pH drops with ultimate pH values typically ranging from 5.3 to 6.0.

Typically, pH of longissimus muscle should show a steady decline to an ultimate pH near 5.5. However, when pork exhibits a subtle pH decline with an ultimate pH above 6.0, it is often referred to as DFD. On the other hand, when pork exhibits a rapid pH decline and a low ultimate pH that is around 5.3, it is often referred to as PSE. The isoelectric point of muscle protein is around 5.3.; when the ultimate pH is near 5.3, it reduces the net charge and decreases myofilament spacing and protein solubility, which contributes to decreased WHC and poor processing yield (Irving et al., 1989; Joo et al., 1999). In research, initial (45 min postmortem) pH is often measured. Within the first 45 min postmortem, the animal's body goes through the most dramatic physical and biochemical changes. The rate at which glycolysis occurs can impact the 45 min pH and quality of a pork carcass. The amount of glycogen present in the muscle after exsanguination is broken down and directly correlated to the amount of lactic acid (Lonergan, 2008). Thus, more glycogen in the muscle results in a lower ultimate pH (Lonergan, 2008). It is reported when 45 min pH is 6.0 or below, the muscle will appear pale and exudative, and when 45 min pH is 6.0 or above, reflectance is little reduced but exudate decreases rapidly (Warriss & Brown, 1987). This suggest that both 45 min and ultimate pH are important in determination of pork quality.

Water-holding Capacity

Water-holding capacity is the ability of meat to retain its moisture content and is affected by biochemical and physical changes which occur postmortem. The WHC of pork is largely influenced by animal stress, genetics, carcass cooling, pH decline, and ultimate pH (Rosenvold & Andersen, 2003; Andres et al., 2007) and is arguably one of the most important quality characteristics of raw products. Water accounts for approximately 75 % of the weight of meat (Schafer et al., 2002); hence WHC is crucial to many meat quality parameters that hold high importance to the industry and consumers (Huff-Lonergan & Lonergan, 2005). The decrease in WHC results in loss of water causing meat to become less juicy and tougher and, thus, less desirable to consumers.

Water within the muscle can be classified into three types by location or mobility, which are *bound water*, *entrapped water,* and *free water*. Bound water is water molecules that are bound to the protein, has reduced mobility, and accounts for a very small fraction of the total water in muscle cells. The amount of bound water changes very little if at all in post-rigor muscle (Offer & Kinght, 1988b). Entrapped water is water that may be held within the structure of muscle but not bound to protein as bound water. Such water is most affected by the rigor process and conversion of muscle to meat as it relies on the structure of muscle. Free water is water that flows freely within the muscle, is held by weak surface forces, and can develop as the muscle structure changes post-rigor. Larger amounts of water such as entrapped water and free water, are often responsible for drip loss in fresh meat as the WHC of meat slowly decreases due to structure change and pH decline.

Traditionally, WHC can be measured as drip loss or ultimately observed as purge in fresh meat packaging. Drip loss can be measured hanging core samples for 24 h at 3 °C. Drip loss is determined by subtracting the end weight from the initial weight and is expressed as a percentage of initial sample weight. A higher drip loss value corresponds to lower WHC. Product weight losses due to purge can average as much as 1-3 % in fresh retail cuts (Offer & Knight, 1988a) and can be as high as 10 % in PSE products (Melody et al., 2004).

Computer Vision System

Computer vision system (CVS) is a system that contains an illumination system, a camera, and image analyzing software and utilizes a personal computer. It has been widely utilized in the food industry and is known to be rapid, economic, consistent, accurate, and non-invasive (Sun, 2000). In the food industry, it has been widely used for different features measurement such as detection or grading to differentiate color, size, texture feature, shape, and uniformity of the product or object (Sun, 2000). Many efforts have been made enhancing the utilization of CVS in the agriculture industry such as classification of types of cereal grains (Luoet al., 1999; Paliwal et al., 2001), color grading for apples (Nakano et al., 1992), and detection of bruises on strawberries (Nagata et al., 2006).

In the beef industry, CVS has been utilized to objectively measure multiple features of beef quality such as marbling and yield percentage using the "beef cam". Research has shown the potential of CVS in predicting beef color (Larraín et al., 2008), fat color, (Chen et al., 2010), tenderness (Li et al., 1999, 2001; Tan, 2004; ElMasry et al., 2012; Sun et al., 2012), pH value (ElMasry et al., 2012), and marbling (Chen et al., 2010; Jackman et al., 2009). Research on the application of CVS in the pork industry is behind other fields with most research occurring within the past decade. Research has focused on the use of CVS for detecting pale, soft, and exudative (PSE) pork (Warriss et al., 2006; Chmiel et al., 2011, 2016), predicting pork color (Lu et al., 2000; Faucitano et al., 2005; Huang et al., 2013; Liu & Ngadi, 2014; Xin et al., 2016), and even for detection of *Escherichia coli* contamination (Tao & Peng, 2014).

Raman Spectroscopy

Raman spectroscopy provides qualitative and quantitative information at the molecular level by measuring the in-elastic light scattering when illuminating the sample with a strong laser. It is a noninvasive spectroscopic technique providing detailed information about the chemical composition of pork. It is also relatively insensitive to water, hence does not suffer from water interference making it a great technology for pork. Disadvantage of using Raman spectroscopy is the size of region of interest. Research utilizing Raman spectroscopy has been able to predict early postmortem and ultimate pH with high accuracy (Scheier, 2015). Raman spectroscopy has also been implemented on determination of fatty acid composition in pork adipose tissue. Results show a high accuracy in predicting the poly- and mono-unsaturated and saturated fatty acids within pork adipose tissue (Berhe, 2016).

Vis/NIRS

Another spectroscopic method is Vis/NIRS analysis, which uses visible light and near-infrared region of the electromagnetic spectrum. It provides an objective, repeatable, accurate method to predict qualitative attributes and chemical composition in meat. Pork was classified into three quality grades (PSE, RFN and DFD) using Vis/NIRS with an overall accuracy of 96 % (Barbin, 2012). It also showed that color reflectance, pH, and drip loss could be predicted with R^2 of 0.93, 0.87, and 0.83, respectively, when using a fresh pork chop. When using frozen pork chops as samples, R^2 for predicting drip loss, color (L*, a*, and b*), and cooking loss were 0.762, 0.906, 0.716, 0.814, and 0.845, respectively (Xie, 2015). Chemical composition traits of protein, moisture, and fat were predicted with R^2 of 0.92, 0.87, and 0.95, respectively (Barbin, 2013).

Hyper Spectral Imaging

Hyper spectral imaging is a combination of spectroscopic techniques and computer image analysis. It shares the advantage of both Raman spectroscopy and CVS, being chemical sensitive and capable of collecting data of the whole surface. However, the enormous amount of data being collected in each hyperspectral cube results in time consuming data collection and data analysis. In 2007, Jun Qiao et al. found, by using wavelengths (430-465 nm & 780-864 nm) and artificial neural network, they could establish a model to successfully group RFN and RSE (red, soft, and exudative) pork with 85 % accuracy. Pork quality traits of drip loss, pH, and color were predicted with correlation coefficients of 0.77, 0.55, and 0.86, respectively. When Gabor filter, image texture feature extraction was applied with hyperspectral imaging, a statistically significant improvement in grouping pork quality (RFN, PFN, PSE, and RSE) was achieved (Liu, 2010). Other attributes such as moisture content of pork chops were successfully predicted with R^2 of 0.94 (Ma, 2017). A safety attribute, *E. coli* contamination, was also studied and predicted with high accuracies (Tao, 2012, 2014). In 2013, Huang et al. predicted total viable count, most important index in evaluation of quality and safety of meat, with a successful R^2 of 0.83. Total volatile base nitrogen content, an indicator of raw meat chemical spoilage, was found to have an R^2 of 0.854 when obtained using salted and cooked pork chop as sample (Cheng, 2017).

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CHAPTER 2. EVALUATION OF PORK MARBLING USING COMPUTER VISION SYSTEM

Abstract

The objective of this study was to investigate the ability of computer vision system (CVS) to predict pork intramuscular fat percentage (IMF%). Center-cut loin samples (n=85) were trimmed of subcutaneous fat and connective tissue. Images were acquired and pixels were segregated to estimate IMF% for each sample. Eighteen image color features were extracted from each image. Subjective marbling scores (SMS) were determined by a trained grader for each image. Crude fat percentage (CF%) was calculated using the ether extract method. Image color features and computer-estimated IMF% were used as predictors for stepwise regression and support vector machine (SVM) models. Results showed that SMS had a correlation of 0.81 with CF% while the computer-estimated IMF% had a 0.66 correlation with CF%. Correlations between computer-estimated IMF% and SMS were 0.62. Accuracy rates for regression models were 0.63 for stepwise and 0.75 for SVM. These results indicate that CVS has the potential to be used as a tool in predicting pork IMF%.

Introduction

It is generally accepted that marbling, or intramuscular fat (IMF), is an important factor which positively influences meat quality. Pork loins with more marbling have been shown to be associated with greater juiciness, flavor, and tenderness scores (Fernandez, Monin, Talmant, Mourot, & Lebret, 1999a; Brewer, Zhu, & McKeith, 2001; Cannata et al., 2010). Fortin, Robertson, and Tong (2005) suggested that once IMF percentage (IMF%) exceeds a threshold level of 1.0 %, it does not affect instrumental measurement of tenderness. The amount of visible fat in loin chops was found to have a negative effect on overall appearance acceptability to consumers because chops with a greater degree of marbling appeared to be more fat and lighter in lean color (Brewer et al., 2001). However, consumer ratings of juiciness, initial tenderness, and flavor were significantly greater ($P < 0.05$) for high IMF pork compared to low IMF pork when cooked, suggesting a discrepancy between visual raw product acceptance and cooked product acceptance (Brewer et al., 2001). In 2010, Moeller et al. reported even though little, an increase of IMF resulted in improvement of pork palatability and in consumer's likelihood of purchase. Japan and Korea desire high quality pork which is dark in color and has a high percentage of IMF (Ngapo, Martin, & Dransfield, 2007; Oh & See, 2012) and account for 50.14 % on a value basis of US pork exports (USMEF, 2015).

Current methods to measure IMF in pork are limited. In laboratory-based research, IMF can be determined by using the crude fat extraction method (AOAC, 1990); however, this extraction method is labor and time intensive. The most common way to determine IMF in the pork industry is by subjective assessment. Assigning subjective marbling scores requires trained individuals assessing the pork loins using pork marbling standard cards (NPB, 2011) on a scale of 1 to 10 (1=devoid, 10=abundant). However, since this is a subjective measurement, variation can occur due to different evaluators, different lean color, and different environmental factors in the slaughter plant such as lighting.

Computer vision system (CVS) is a system that contains an illumination system, a camera, and image analyzing software utilizing a computer. It has been widely utilized in the food industry and is known to be rapid, economic, consistent, accurate, and non-invasive (Sun, 2000). Many efforts have been made enhancing the utilization of CVS in the agriculture industry such as classification of types of cereal grains (Luo, Jayas, & Symons, 1999; Paliwal, Visen, &

Jayas, 2001), color grading for apples (Nakano, Kurata, Kaneko, Kazama, & Takizawa, 1992), and detection of bruises on strawberries (Nagata, Tallada, & Kobayashi, 2006).

In the beef industry, CVS has been utilized to objectively measure multiple features of beef quality such as marbling and yield percentage using the "beef cam" (Cannell et al., 2002). Research has shown the potential of CVS in predicting beef color (Larraín, Schaefer, & Reed, 2008), fat color, (Chen, Sun, Qin, & Tang., 2010), tenderness (Li, Tan, Martz, & Heymann, 1999; Li, Tan, & Shatadal, 2001; Tan, 2004; Sun et al., 2012; ElMasry, Sun, & Allen, 2012), pH value (ElMasry et al., 2012), and marbling (Chen et al., 2010; Jackman, Sun, & Allen, 2009). Research on the application of CVS in the pork industry has further developed in the past decade. Research has focused on the use of CVS for detecting pale, soft, and exudative (PSE) pork (Warriss, Brown, & Paściak, 2006; Chmiel, Słowiński, & Dasiewicz, 2011; Chmiel, Słowiński, Dasiewicz, & Florowski, 2016), predicting pork color (Lu, Tan, Shatadal, & Gerrard, 2000; Faucitano, Huff, Teuscher, Gariepy, & Wegner, 2005; Huang, Liu, Ngadi, & Gariépy, 2013; Liu & Ngadi, 2014; Xin et al., 2016), and even for detection of *Escherichia coli* contamination (Tao & Peng, 2014).

Support vector machine (SVM) is a novel machine learning technology which has been widely utilized in food or agriculture industry for classification problems such as olive oil (Devos, Downey, & Duponchel, 2014), milk (Brudzewski, Osowski, & Markiewicz, 2004), rice (Kaur & Singh, 2013), pizza sauce spread (Du & Sun, 2005), fish filet (He, Wu, & Sun, 2014), white vinegar (Bao et al., 2014). It is a commonly used pattern recognition algorithm which has been proven to be robust and accurate. In our research SVM is an appropriate method for multivariate data image based research (Sun, Chen, Berg, & Magolski, 2011; Sun et al., 2012; Sun et al., 2014; Sun et al., 2016)).

However, little research of using CVS as a tool for objective measurement of IMF% in pork loin chops has been conducted (Faucitano et al., 2005; Huang et al., 2013). Thus, the objective of this research was to evaluate the potential of using CVS to estimate IMF% in pork and to set up IMF% prediction models using stepwise regression and SVM modeling. Furthermore, the objective of this research is to compare the accuracy of our CVS system to subjective IMF% determined by a trained grader.

Material and Methods

Pork Sample Preparation

Boneless center-cut pork loin sections (10.16 cm thick, n = 85) were obtained from two slaughter facilities. Samples were shipped overnight to North Dakota State University (NDSU). After arrival at NDSU, a 2.54-cm thick chop was taken from the center of the loin sample. Chops were then trimmed of subcutaneous, intermuscular fat, and connective tissue to be used for computer imaging and ether extract analysis for IMF%.

Color Image Acquisition

After trimming, an image was acquired on both sides of the samples using the computer vision system. The computer vision system (Fig. 2.1) consists of three components: a three charge-coupled device color digital camera (Model S2100HD, Fujifilm Corporation, Japan) with supporting lighting system consisting of two white LED bar lights (Lux = 401; YX-BL25040, Yongxin Ltd., China), and a personal computer (850 MHz AMD Athlon processor with 1024 MB RAM). Images were processed and analyzed using Matlab software (Version 7; The Math-works, Natick, MA, USA). A self-designed image acquisition studio with light-absorbent black fabric background was established in order to provide a controlled and consistent lighting environment. A dome-shaped reflective polyethylene material was installed to assist in the even

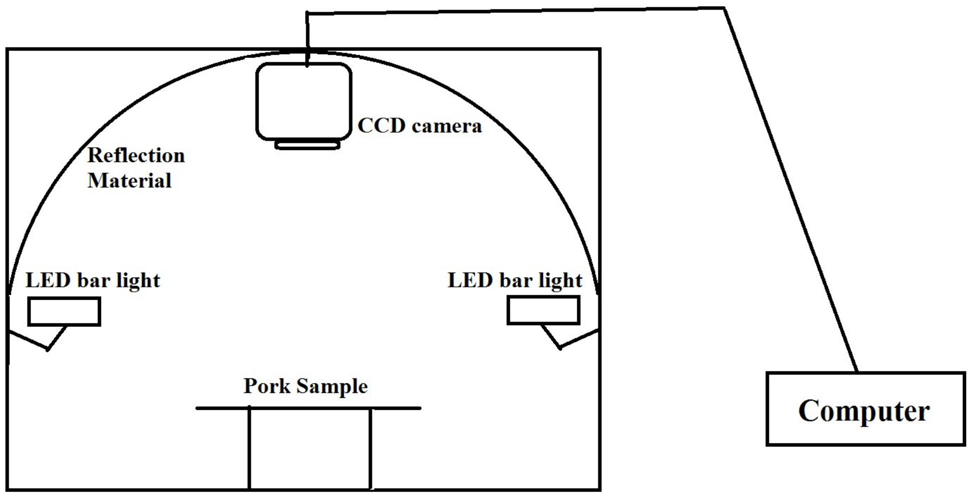

Figure 2.1. Computer vision acquisition system. *CCD = charge coupled device.

distribution of light from the bar lights inside the studio. The RGB (red, green, blue) setting for camera was calibrated using a white plate before each period of image acquisition.

Image Processing and Color Features Extraction

The original image acquired by the computer vision system is shown in Fig. 2.2(a). Using the Matlab software, the background of the image was removed using the gray level histogram Otsu method (Otsu, 1975; Fig. 2.2(b)). After removing the background, the image was binarized

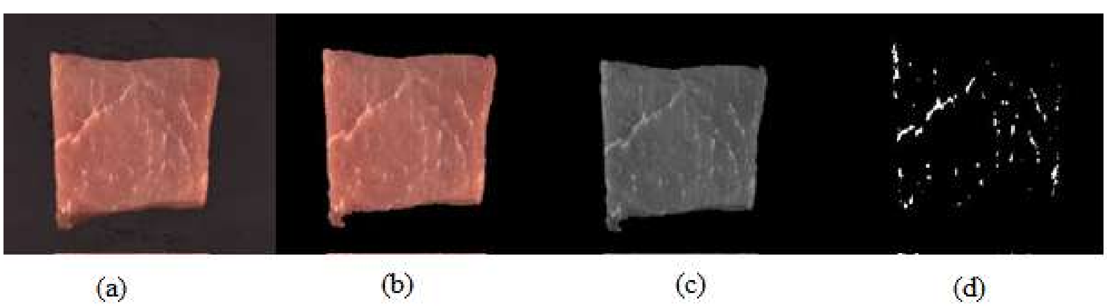

Figure 2.2. Segmentation procedure: (a) Original pork sample image. (b) Removal of background. (c) B channel shown in gray scale for lean and fat segmentation. (d) Fat segmentation.

on the optimum threshold value according to the color space blue (B) layer (Fig. 2.2(c)) which was done by calculating the B value of every pixel and automatically estimating the threshold between lean and fat tissue (Fig. 2.2(d)) according to Sun's method (Sun et al., 2014).

The number of fat and lean tissue pixels was calculated by computer pixel counting method using images (both sides) from each sample. Total pixels of the sample were the sum of fat and lean tissue pixels. IMF% from image processing method was estimated as below:

Image IMF% = Marbling area pixel / (Marbling area pixel + Lean area pixel) (Eq. 2.1)

Eighteen image color components (the mean, μ, and standard deviation, σ, of each color space panel) were extracted from three color spaces: RGB (red, green, blue), HSI (hue, saturation, intensity), and L*a*b* (black to white, green to red, blue to yellow) according Sun's method (Sun, 2016).

Subjective Marbling Score & Lipid Extraction

Subjective marbling score (SMS) was determined by a trained evaluator using pork quality standard cards (NPB, 2011) based on the original images. A grade of marbling was giving from the range of 1 (devoid of marbling) to 3 (moderate amount of marbling). The SMS was determined and averaged based on both sides images from a sample.

After images were acquired, samples were freeze dried for 48 h to remove moisture. After the freeze drying period, crude fat percentage (CF%) was determined gravimetrically using Soxhlet extraction procedure with petroleum ether (AOAC, 1990).

Statistical Analysis

The means procedure in SAS (v. 9.4; SAS Institute, Inc., Cary, NC, USA) was used to estimate the simple statistics for CF%, SMS, and image IMF%. The correlation procedure in

SAS was used to estimate the Pearson correlations between CF%, SMS, and image IMF%. Simple statistics for the 18 color features were also estimated using the means procedure in SAS.

Stepwise regression models were constructed by recursively adding or deleting one independent predictor at a time. Stepwise regression modelling will include the independent predictor with the lowest P-value provided it meets the threshold for entrance into the model and then will check to ensure all predictors in the model meet the threshold for retention in the model, removing one predictor at a time if it does not meet the threshold for retention in the model. This continued until the same variable was being added and removed or no variables were added or removed. In this study, the reg procedure in SAS was utilized to conduct the stepwise regression analysis, using a significance level of $P < 0.15$ for entrance into the prediction model and $P < 0.10$ for retention in the prediction model.

In order to test and improve the stability and robustness of the model, the bootstrap method by Efron (1979) was adopted using SAS. Using the image IMF% value and the 18 color features, stepwise regression modelling was conducted using the bootstrap technique. Data was divided into training (70 %) and test (30 %) data sets. After 100 repetitions, a scree test was conducted to determine which variables to include in the final model based on the number of times they were included in the stepwise regression model. After obtaining the variables of interest for the final model, the bootstrap technique was utilized again for stepwise regression. Again splitting the data into training (70 %) and test (30 %) and running 1000 replications, a final model was developed. The final model was then used to estimate the IMF% (RIMF%) and compared to the actual IMF% from the ether extraction.

Support vector machine (SVM) proposed by Cortes and Vapnik (1995) is a supervised learning technique with associated learning algorithms which is widely used for classification and regression problems. The SVM can effectively perform binary non-linear classification by using the kernel trick, where it calculates the hyperplane which maximizes the distance to the closest samples of both classes. Therefore, it is very important to choose the appropriate kernel function before constructing an SVM classifier. Currently, the popular kernel functions include polynomial darnel function, sigmoid kernel function, and radial basis function (RBF) kernel function. Howley and Madden (2005) has shown that RBF kernel function performs best and is widely used in SVM; therefore, in this study, the RBF kernel function was used for SVM. The RBF kernel function can be written as:

$$K(x_i y_i)=\exp(-y\|x_i-x_j\|^2) \qquad \text{(Eq. 2.2)}$$

where $x \in Rn$ is n-dimension vector and $y_i \in \{-1, +1\}$ is the class label, γ is a parameter which should be specified by the model user. More details regarding selecting the appropriate kernel parameter, γ, and penalty constant, C, can be found in Sun et al. (2014). A multiclass SVM classifier was conducted since the SMS grade ranged from 1 to 3. Multi-SVM classifier can be developed by combining two class SVM classifiers using one of two strategies: one-versus-one or one-versus-rest (Herbrich, 2004). In this study, a one-versus-one method was adopted and the data was divided into a training set and a test set (approximately 70/30 split) for SVM model validation.

Two methods were used to determine accuracies of the stepwise regression and SVM models. The first was to calculate the residual (CF% - RIMF%). If the absolute value of the residual was less than 0.5, then the estimate was considered to be correct. The second was to give each sample a categorical value based on the CF% and the RIMF%. The categorical values were 1 (IMF% < 2), 2 (2 ≤ IMF% < 3), and 3 (IMF% ≥ 3). After categorical values were assigned, the percent classified correctly per original category and overall were calculated.

Results and Discussion

Simple Statistics and Correlations

Simple statistics of the different methods of obtaining IMF% is shown in Table 2.1. The SMS is less variable and greater on average than CF%. Image IMF% is more variable and lower on average than CF%. Pearson correlation coefficients between CF%, image IMF%, and SMS are reported in Table 2.2. Simple statistics for the 18 color features extracted are reported in Table 2.3.

Table 2.1. Simple statistics for three different methods of estimating intramuscular fat percentage (IMF%).

Method	N	Mean	S.D.	Minimum	Maximum
Ether Extract IMF%	85	2.13	0.90	0.60	4.33
Subjective IMF%	85	2.38	0.65	1	4
Image IMF%	85	1.91	1.31	0.21	7.01

Table 2.2. Pearson correlation coefficients between three different methods of estimating intramuscular fat percentage (IMF%).

	Image IMF%	Subjective IMF%
Ether Extract IMF%	0.62*	0.81*
Image IMF%		0.66*

* Correlations are significant at $P < 0.0001$.

In the current study, the Pearson correlation coefficient between image IMF% (calculated by pixels) and SMS was 0.66. Huang et al. (2013) had similar results using grey-level co-occurrence matrix (GLCM) based and wide line detector (WLD) methods ($r^2 = 0.79$ and 0.94, respectively). Additionally, the Pearson correlation between image IMF% and CF% was 0.66 in the current study which was consistent with the result ($r^2 = 0.60$) obtained by Faucitano et al. (2005). These results suggest a strong relationship with computer obtained marbling values with SMS and actual IMF%.

Table 2.3. Simple statistics for image color features.

Color feature[1]	N	Mean	S.D.	Minimum	Maximum
μ_R	85	157.88	19.39	112.87	192.14
μ_G	85	98.05	12.75	66.86	122.86
μ_B	85	88.13	11.27	61.42	109.53
σ_R	85	31.61	5.86	18.20	43.33
σ_G	85	20.07	3.68	11.66	28.58
σ_B	85	18.23	3.32	10.69	25.52
μ_H	85	6.20	1.62	3.82	10.50
μ_S	85	24.91	6.48	15.37	42.11
μ_I	85	114.69	14.39	80.38	140.70
σ_H	85	0.05	0.01	0.03	0.06
σ_S	85	0.15	0.02	0.11	0.18
σ_I	85	23.22	4.26	13.44	32.46
μ_{L^*}	85	71.17	3.84	61.27	77.84
μ_{a^*}	85	13.59	1.00	11.51	16.12
μ_{b^*}	85	9.79	0.58	8.31	11.28
σ_{L^*}	85	14.13	1.93	9.59	18.02
σ_{a^*}	85	2.74	0.43	1.94	3.61
σ_{b^*}	85	2.00	0.28	1.43	2.53

[1] Mean, μ, and standard deviation, σ, for each image for R (red), G (green), B (blue), H (hue), S (saturation), I (intensity), L* (black to white), b* (blue to yellow), and a* (green to red).

Stepwise Regression

After the initial bootstrapping (100 repetitions with all 18 color features and image IMF% included to be fit in the stepwise regression model), the scree test (Fig. 2.3) was used to determine the variables to retain in the final model. Based on the scree test, Image IMF%, μ_{a^*}, and μ_{L^*} were retained.

After rerunning the bootstrapping method with 1000 repetitions and only image IMF%, μ_{a^*}, and μ_{L^*} included, image IMF% and μ_{a^*} were included in every run while μ_{L^*} was only included 26.3 % of the time. Simple statistics from the bootstrapping stepwise regression model are presented in Table 2.4. Therefore, the final model for stepwise regression analysis was:

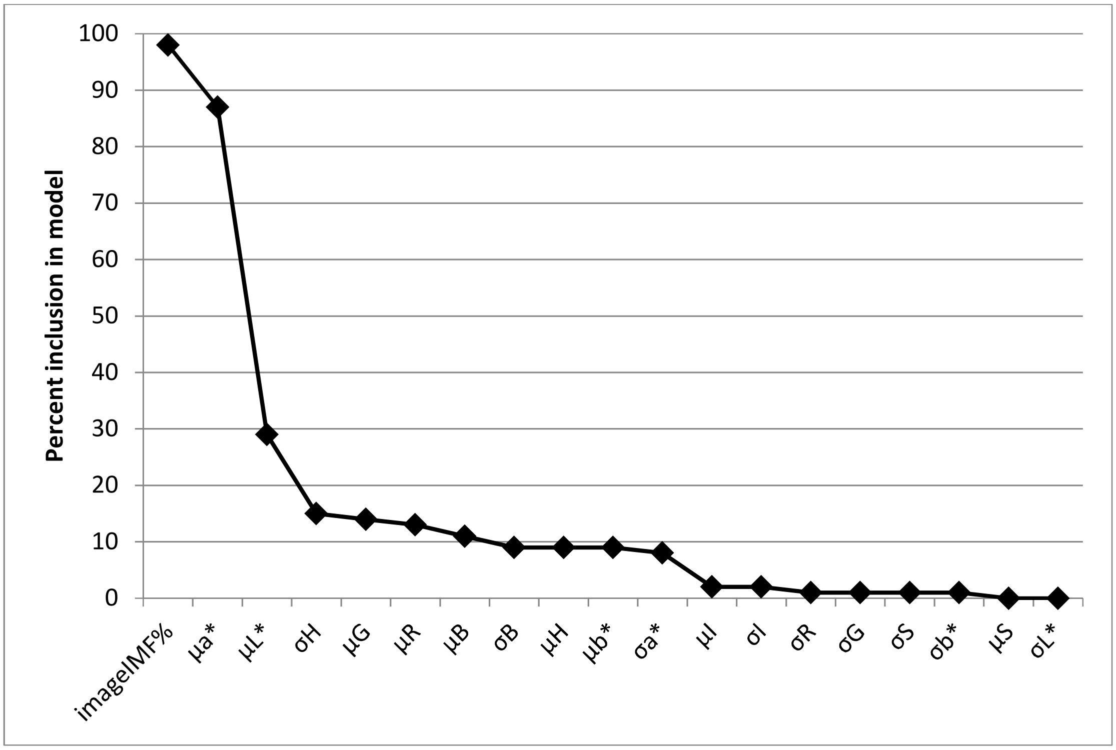

Figure 2.3. Scree test results for inclusion in model for Image intramuscular fat percentage (IMF%) and mean, μ, and standard deviation, μ, of 18 color features: R (red), G (green), B (blue), H (hue), S (saturation), I (intensity), L* (black to white), a* (green to red), and b* (blue to yellow).

Table 2.4. Simple statistics for bootstrap resampling of stepwise regression analysis.

Variable	N	Mean	S.D.	Minimum	Maximum
Intercept	1000	7.17	1.26	4.94	10.59
β Image IMF%[a]	1000	0.53	0.04	0.44	0.72
β μ_{L*}[b]	1000	-0.01	0.02	-0.07	0
β μ_{a*}[c]	1000	-0.38	0.04	-0.53	-0.21
R^2	1000	0.55	0.04	0.40	0.70

[a] Regression coefficient, β, for Image intramuscular fat percentage (IMF%).

[b] Regression coefficient, β, for the mean, μ, value for L* (black to white).

[c] Regression coefficient, β, for the mean, μ, value for a* (greed to red).

$$\text{Estimated IMF\% (RIMF\%)} = 7.1732411 + 0.5306245 \times \text{Image IMF\%} - 0.0117744 \times \mu_{L*} - 0.3827521 \times \mu_{a*} \quad \text{(Eq. 2.3)}$$

With the final model, an RIMF% was calculated for each of the 85 samples in the original dataset. A residual (IMFresid) was calculated for each observation as CF% minus RIMF%. The distribution of residuals is presented in Figure 2.4. The accuracy of the model was determined as the percentage of observations falling within 0.5 of the CF% value. The accuracy of the final model was determined to be 65.1 % with an additional 23.3 % of samples having an estimated IMF% within 1 of the CF% value. No samples had an RIMF% deviated greater than 2 from the CF%. The accuracies based on categories 1, 2, and 3 are presented in Figure 2.5. For category 1, 76.2 % were classified as 1 and 23.8 % as 2. For category 2, 68.0 % were classified correctly and 16.0 % were classified as 1 and 16.0 % as 3. For category 3, only 31.6 % were classified

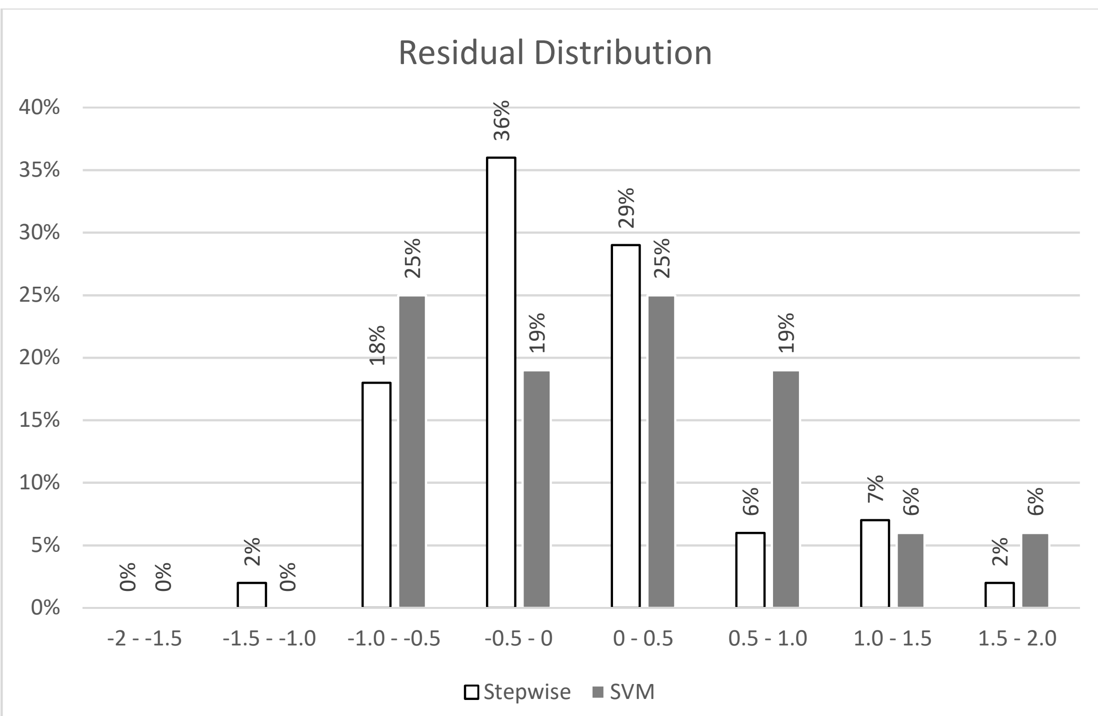

Figure 2.4. Distribution of residuals from stepwise regression and support vector machine (SVM) models

correctly with 52.6 % being classified as 2 and 15.8 % as 1. Overall, 63.9 % were classified correctly, 32.6 % were classified 1 category off, and 3.5 % were classified 2 categories off.

SVM Model Prediction from Color Images

The SVM modelling only reports the estimate of the samples in the test data set. Therefore, there is no equation to report. Figure 2.4 shows the distribution of residuals. The accuracy of the model was determined as the percentage of observations falling within 0.5 of the CF% value. The accuracy of the final model was determined to be 65.0 % with an additional 35.0 % of samples having an estimated IMF% within 1 of the CF% value. No samples had an estimated IMF% deviation greater than 2 from the CF%. The accuracies based on categories 1, 2, and 3 are presented in Figure 2.5. For category 1, there was only 1 sample in the test data set and it was classified as a 2. For category 2, 80 % were classified correctly and 10 % were classified as 1 and 10 % as 3. For category 3, 80 % were classified correctly with 20 % being classified as

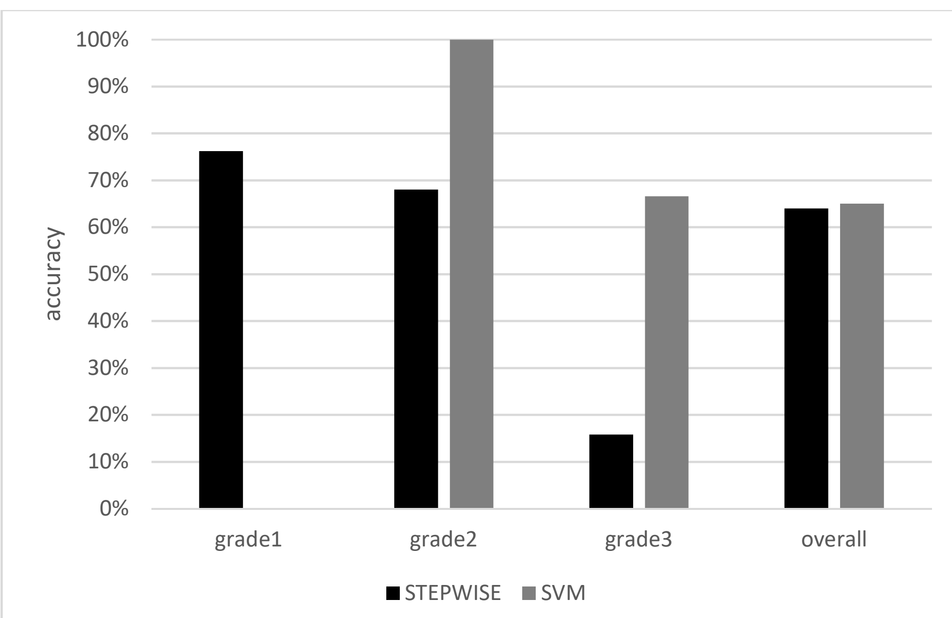

Figure 2.5. Comparison of stepwise model and SVM model to predict pork IMF% using image pixel area values and color features

2 and 0 % as 1. Overall, 65 % were classified correctly, 35 % were classified 1 category off, and 0 % were classified 2 categories off.

Overall Comparison of the Stepwise Linear Regression and SVM Classifier

In this study, stepwise regression was compared with SVM regression. There was a noticeable difference in the overall accuracy and the prediction residuals. Many factors contributed to such result. In the stepwise model while 18 color features were extracted from the image, after running bootstrap method and scree test, only image IMF%, μ_{L*}, and μ_{a*} were chosen to remain in the final formula. All samples (85) were used to test the accuracy of the model. The SVM regression is a machine-learning technique which can be trained to use all predictors including meaningful or nontrivial relationships utilizing training data and then use these relationships from the data base to predict new, unlabeled test data unlike stepwise regression which fits in suitable predictors and eliminate useless ones. Therefore, all 18 color features and image IMF% were used in the SVM model. However, the bootstrap method was not utilized for SVM model; hence only 30% (16) of the samples were used to test the model. Meanwhile the difference between sub-set and re-sampling may play a huge factor in establishment of the model and result of accuracy due to the size of sample.

The distribution of the residuals from both models is shown in Figure 2.4. The SVM model has a higher prediction accuracy result; however, the distribution of residuals shows that stepwise model has a more favorable residual distribution. Also from the residual distributions, it shows that the SVM model will have a higher estimate than the actual result while the stepwise model, even though more evenly distributed, tends to underestimate the CF%. This might be due to the difference between the factors that were used to setup the model or the fundamental algorithm of the two models.

Stepwise regression model predicted CF% correctly approximately 64 % of the time while the SVM model predicted correctly 75 % of the time. While the accuracies are not extremely high, they show potential for developing a model to predict CF%. Several factors were considered to modify in order to improve the predict accuracy, such as number of samples needs to be expanded. With a greater number of samples in the training dataset, there would be better representation of samples along the entire range of CF%, resulting in less extrapolation of the data. For example, the randomization of creating the training and testing datasets for the SVM model resulted in the testing dataset only having one sample of grade 1. With a different subsampling, the training dataset could have only had one sample of grade 1. With a larger dataset, the odds of being limited on samples of a particular grade would be decreased. A larger dataset could also result in a better model being developed which could increase accuracy. It is difficult for the CVS to detect very fine marbling. More research involving advanced image analyzing techniques and cameras which take pictures of higher quality and clarity needs to be conducted and would be beneficial in the development of an automated objective method of measuring IMF%.

Conclusions

In this study, stepwise regression and SVM modelling were utilized to predict IMF% in pork samples. The results demonstrate the potential of using CVS and color images of pork samples to estimate IMF%. The distribution of residuals is promising for the use of CVS as a tool to predict IMF% in pork in the future. Further work will be to expand sample size through all categories of IMF%, building proper industrial implementation for marbling evaluation, and exploring potential image analysis techniques to improve distinction of fine marbling in pork.

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CHAPTER 3. PREDICTING PORK CHOP INTRAMUSCULAR FAT USING VISION SYSTEM TECHNOLOGY

Abstract

The objective of this study was to examine the potential of using computer vision system (CVS) as a tool to predict pork chop intramuscular fat (IMF) percentage under industry scale equipment and environment. An anterior (3rd rib) and posterior (10th rib) chop were obtained from 200 pork loins from 7 different packing plants (n = 2800 chops). Color images of pork chop samples were acquired using a CVS. Subjective marbling scores (SMS) were determined according to the National Pork Board standards (NPB, 2011) by a trained evaluator. Crude fat percentage (CF%) was calculated using ether extract method (AOAC, 1990). Results show that SMS had an overall accuracy of 70.1 % for predicting CF%, while accuracies were 90.0, 44.5, 34.0, 30.4, and 44.1 % for CF% categories of 0-1.99, 2-2.99, 3-3.99, 4-4.99, and greater than 5 %, respectively. Comparatively, the overall accuracy of using CVS was 68.6 %, with individual accuracies being 78.6, 48.5, 23.7, 13.3, and 24.4 % for CF% categories of 0-1.99, 2-2.99, 3-3.99, 4-4.99, and greater than 5 %, respectively. These results demonstrate the potential of using CVS as an objective measurement for pork chop CF% within the industry environment.

Introduction

It is generally accepted that marbling, or intramuscular fat (IMF), is an important factor which positively influences meat quality. Pork loins with more marbling have been shown to be associated with greater juiciness, flavor, and tenderness scores (Fernandez, Monin, Talmant, Mourot, & Lebret, 1999a; Brewer, Zhu, & McKeith, 2001; Cannata et al., 2010). Fortin, Robertson, and Tong (2005) suggested that once IMF percentage (IMF%) exceeds a threshold level of 1.0 %, it does not affect instrumental measurement of tenderness.

Consumers from different culture have different preference in pork quality traits such as color and marbling. While the amount of visible fat in loin chops was found to have a negative effect on overall appearance in the U.S. (Brewer et al., 2001), it is reported that consumers from Japan, Korea find well marbled pork more appealing whereas consumers from Taiwan, Poland and Australia prefer lean pork (Ngapo et al., 2013). When cooked, consumer ratings of juiciness, initial tenderness, and flavor were significantly greater ($P < 0.05$) for high IMF pork compared to low IMF pork, regardless of visual preference of the raw product (Brewer et al., 2001). Japan and Korea desire high quality pork which is dark in color and has a high percentage of IMF (Ngapo, Martin, & Dransfield, 2007; Oh & See, 2012) and account for 50.14 % on a value basis of U.S. pork exports (USMEF, 2015).

Current methods to measure IMF in pork are limited. In laboratory-based research, IMF can be determined by using crude fat extraction (AOAC, 1990); however, this extraction method is labor and time intensive. The most common way to determine IMF in the pork industry is by subjective assessment. Assigning subjective marbling scores requires trained individuals assessing the pork loins using pork marbling standard cards (NPB, 2011) on a scale of 1 to 10 (1=devoid, 10=abundant). However, since this is a subjective measurement, variation can occur due to different evaluators, different lean color, and different environmental factors in the slaughter plant such as lighting, fatigue or exhaustion of eye sight, and the repeatability is poor. This suggest that a rapid, accurate, repeatable, objective measurement to determine IMF in pork is still needed.

Computer vision system (CVS) is a system that contains an illumination system, a camera, and image analyzing software utilizing a computer. It has been widely utilized in the food industry and is known to be rapid, economic, consistent, accurate, and non-invasive (Sun, 2000).

In the beef industry, CVS has been utilized to objectively measure multiple features of beef quality such as marbling and yield percentage using the “beef cam” (Cannell et al., 2002). Research has shown the potential of CVS in predicting beef color (Larraín, Schaefer, & Reed, 2008), fat color, (Chen, Sun, Qin, & Tang., 2010), tenderness (Li, Tan, Martz, & Heymann, 1999; Li, Tan, & Shatadal, 2001; Tan, 2004; Sun et al., 2012; ElMasry, Sun, & Allen, 2012), pH value (ElMasry et al., 2012), and marbling (Chen et al., 2010; Jackman, Sun, & Allen, 2009). Research on the application of CVS in the pork industry has further developed in the past decade. Research has focused on the use of CVS for detecting pale, soft, and exudative (PSE) pork (Warriss, Brown, & Paściak, 2006; Chmiel, Słowiński, & Dasiewicz, 2011; Chmiel, Słowiński, Dasiewicz, & Florowski, 2016), predicting pork color (Lu, Tan, Shatadal, & Gerrard, 2000; Faucitano, Huff, Teuscher, Gariepy, & Wegner, 2005; Huang, Liu, Ngadi, & Gariépy, 2013; Liu & Ngadi, 2014; Xin et al., 2016), and even for detection of *E. coli* contamination (Tao & Peng, 2014).

In 2014 Huang reported that different sampling locations including last rib, 10th rib, 3rd/4th last ribs and 2nd/3rd last ribs have been adopted in different studies, and that the nonuniformity of sampling site could cause discrepancy in results of pork quality studies.

Thus, the objective of this research was to compare the accuracy of using CVS estimated IMF% and subjective marbling score with CF%. Furthermore, to understand if site of sampling would play as an factor to the accuracy of both methods.

Material and Methods

Sample Collection and Image Acquisition

Whole, boneless loin samples were obtained from 7 different processing plants (n = 200 per plant). Each sample was selected by a trained evaluator from the deboning line. Samples were chosen to maximize the variation in pork quality for subjective color and marbling scores, which were assessed on-line according to National Pork Board (NPB) standards (NPB, 2011). After in-plant data collection (data not presented), whole loins were vacuum-packaged and transported in a refrigerated truck to the US Meat Animal Research Center in Clay Center, NE. Whole loins were aged for 14 d and then sliced into individual chops. The chop from the 3^{rd} and 10^{th} ribs were selected and a digital color image was taken of each chop using a computer vision system (CVS) after a 15 min bloom period (Fig. 3.1), consisting of an industry camera (NI 1776C smart camera, National Instrument, Ltd., USA) with a 1/1.8" F1.6/4.4-11-mm lens (LMVZ4411, Kowa, Ltd., Japan), a 44-inch dome light (DL180, advance illumination, Ltd., USA), and a personal laptop (Lenovo, Ltd., China). The CVS was attached to a table to ease transportation of the dome light and to standardize the relationship of the camera to the dome light and the samples. A black, light-absorbent fabric was installed between the dome light and table to exclude light noise from the surrounding environment. Before sample collection, a Minolta white tile was used for calibration. The white tile was placed in the center and corner of the CVS to ensure the evenness of light spread. When taking pictures of the white tile, color space red green and blue color features were extracted and used as standards for calibration and setting of the CVS. Each sample was manually placed on a light-absorbing, black background surface for image acquisition. The color image was captured and stored using LabVIEW software (National Instrument, Ltd, TX). After imaging, samples were covered with plastic wrap and allowed to bloom until 3 h post-cut time. After 3 h, subjective marbling scores (SMS) were assessed by a trained evaluator using the NPB standards. After SMS were assessed, chops were vacuum packaged and transported to North Dakota State University on ice. Once at North Dakota State University, chops were trimmed of connective tissue and subcutaneous fat. Samples were freeze-dried for 48 h to remove moisture. After the freeze-drying period, crude fat percentage (CF%) was determined gravimetrically using Soxhlet extraction with petroleum ether according to AOAC procedure (AOAC, 1990).

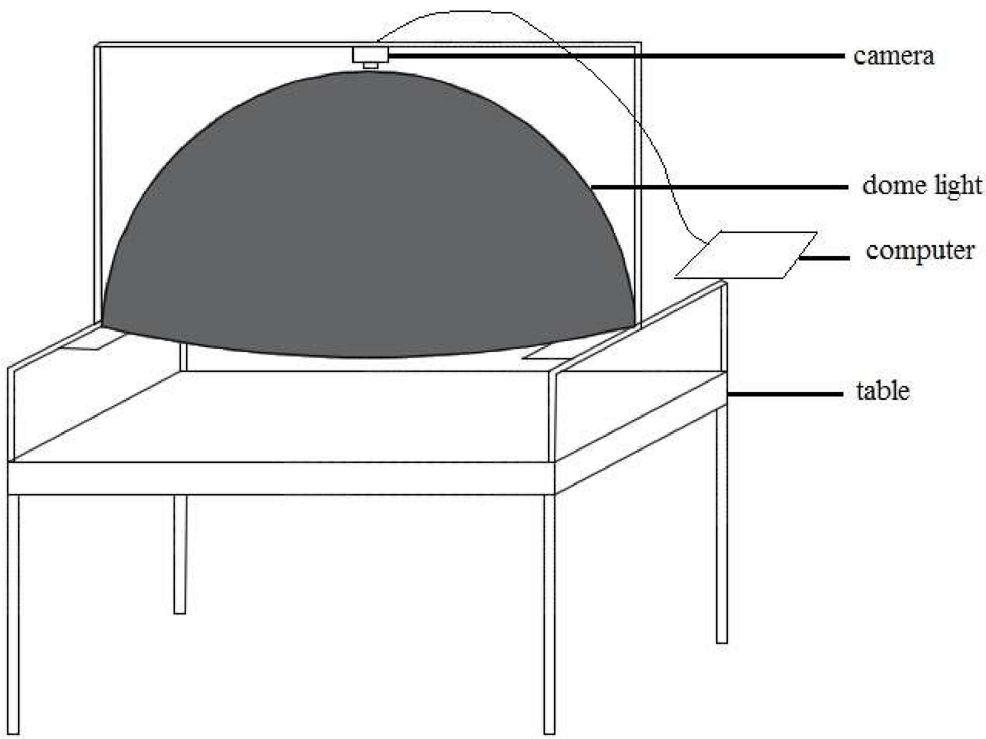
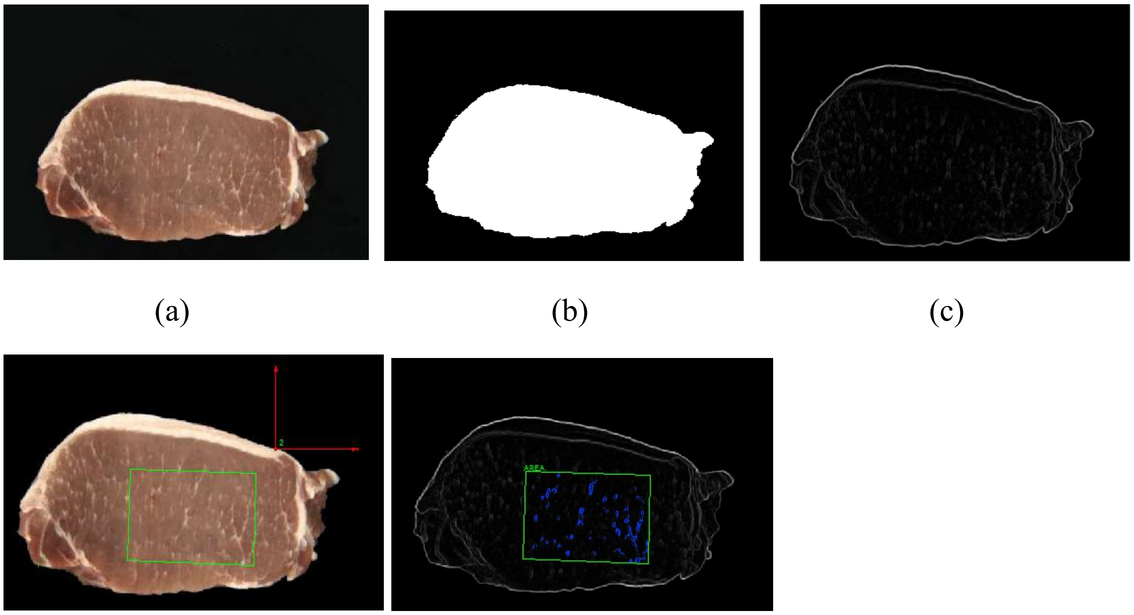


Figure 3.1. Pork color image acquisition system

Image Analysis

An original pork sample image acquired by the CVS is shown in Figure 3.2(a). To remove the background of the image automatically, Otsu method was performed using the LabVIEW software (Fig. 3.2(b); Otsu, 1975). Once the background was removed, the Sobel



(a) (b) (c)

(d) (e)

Figure 3.2. (a) Original image; (b) background segmentation; (c) identification of fat pixels; (d) identification of region of interest; (e) identification of fat and lean muscle pixels within region of interest

Method was applied to the image to segment lean muscle pixels and IMF pixels (Fig. 3.2(c)). The region of interest (ROI; 4.08 × 5.08 cm) was then determined automatically using a mapping system to avoid uncleaned surface or connective tissue remained on the pork loin chop (Fig. 3.2(d)). After determination of ROI, the fat and lean pixels were then counted to calculate image IMF% (IIMF) within ROI (Fig. 3.2(e)).

Data Analysis

In order to calculate the accuracies of SMS predicting CF%, CF% was categorized into ranges of 0-1.99, 2.00-2.99, 3.00-3.99, 4.00-4.99, and greater than 5.00 %, identified as CF1, CF2, CF3, CF4, and CF5, respectively. Bootstrapping (Efron, 1979) was utilized to run 100 replications of randomly dividing the data into training (70 %) and test (30 %) datasets. The training dataset was used to generate a simple regression model of CF% = intercept + β × IIMF. The test dataset was used to determine the accuracy of the equation developed from the training dataset. Intercepts and betas from all 100 replicates were averaged to create a final regression equation to calculate a regression IMF% (RIMF). Accuracies were averaged across all 100 replicates and compared to results from the final regression equation using all data. Since results were similar, results from the final regression equation will be reported. To compare results with SMS, RIMF was also categorized into RIMF1, RIMF2, RIMF3, RIMF4, and RIMF5, using the same values as CF%. Residual distributions of IIMF and RIMF were calculated by subtracting them from CF% to further understand the predictive power of IIMF and RIMF. All procedures were accomplished using SAS (v. 9.4, SAS Institute, Inc., Cary, NC).

Results and Discussions

Distributions

From the 2800 chops that were collected, the distribution of CF% was 1657, 787, 253, 69, and 34 in CF1, CF2, CF3, CF4, and CF5, respectively. By definition of SMS, there should be 59.2, 28.1, 9.1, 2.4, and 1.2 % scored as a 1, 2, 3, 4, and greater or equal to 5, respectively. This distribution of CF%, with 87.3 % of chops having a CF% less than 3, is similar to results from Newman (2015), who found 87% of pork loin chops in the US retail market had a SMS equal or less than 3% marbling.

Comparatively, actual SMS had a distribution of 1840, 611, 208, 98, and 43 for scores 1, 2, 3, 4, and 5 or above, respectively, which corresponds to percentages of 65.7, 21.8, 7.4, 3.5, and 1.5 %, respectively. Three regression models were developed to estimate RIMF, one each for overall, just anterior chops, and just posterior chops. The equations were as follows:

$$\text{RIMF\%} = 1.4641932 + 0.394858972*\text{IIMF\%} \quad \text{(Eq. 3.1)}$$

$$\text{RIMF\%} = 1.624465895 + 0.427090402*\text{IIMF\%} \quad \text{(Eq. 3.2)}$$

$$\text{RIMF\%} = 1.309720432 + 0.357723038*\text{IIMF\%} \quad \text{(Eq. 3.3)}$$

where Eq. 3.1 is for overall, Eq. 3.2 is for anterior chops, and Eq. 3.3 is for posterior chops. The distribution of RIMF for both chops combined was 1838, 774, 149, 33, and 6 for RIMF1, RIMF2, RIMF3, RIMF4, and RIMF5, respectively, which corresponds to percentages of 65.6, 27.6, 5.3, 1.2, and 0.2 %, respectively. The distribution of SMS had more chops in the extreme categories (1, 4, and 5) than CF% while the distribution of RIMF had more chops in the lower categories (1 and 2) than CF%.

Accuracies

When comparing CF% with SMS, the overall prediction accuracy was 70.1 %. When evaluating different levels of marbling, SMS had an accuracy of 90.0, 44.5, 34.0, 30.4, and 44.1 % when predicting CF1, CF2, CF3, CF4, and CF5, respectively (shown in Fig. 3.3). SMS

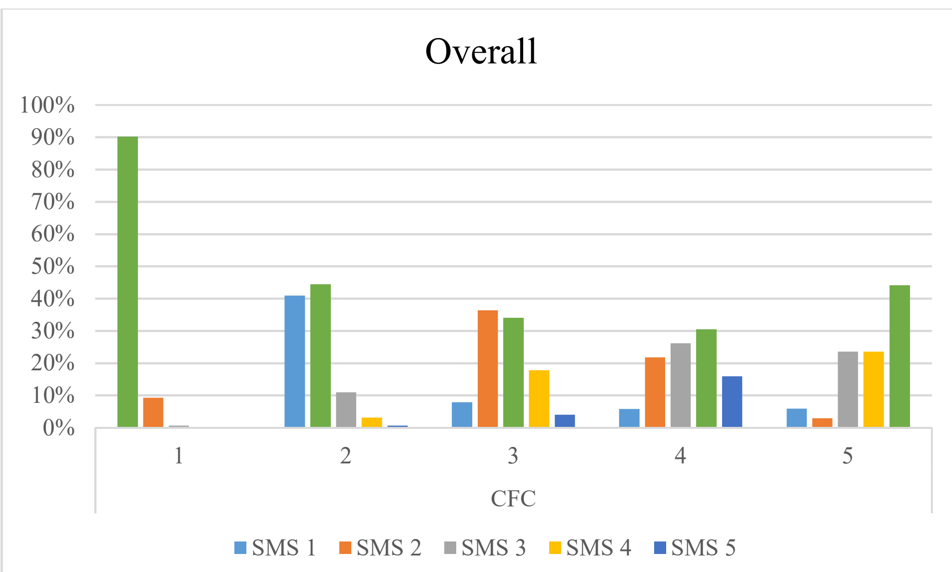


Figure 3.3. Overall crude fat categorical versus subjective marbling scores. Within a crude fat category, the distribution is what the samples were called subjectively and the percentages will sum to 100. The green column represents the accuracy of calling subjective marbling score in each category, whereas light blue, orange, gray, yellow, and dark blue is what the samples were assessed.

had the best accuracy in predicting CF% under 2, whereas when predicting CF% at a higher percentage the accuracies were lowered. In fact, Fig. 3.3 shows that SMS often underestimates CF% at the higher CF%. This could be explained by several factors. Variety in pork appearance can affect SMS. While the NPB standards provide an example for each SMS that corresponds to the CF%, pork chops being assessed may have several different traits that affect the evaluator's ability to correctly assess the CF%, such as different color, texture, exudative, and the fineness or coarseness of IMF. Chops that are paler, more exudative, or have finer IMF are harder for the evaluator to distinguish IMF from the lean tissue. Additionally, evaluator fatigue could contribute to a decreased accuracy of evaluating SMS. When evaluating a large number of samples, the evaluator may become fatigued and be less accurate in calling SMS as an estimate of CF%. These results suggest that, even for trained evaluators, it is hard to subjectively and accurately predict the IMF% of pork chops, particularly when CF% is higher than 2 %. When dividing the pork chop into anterior (3rd rib) and posterior (10th rib), similar results were found. The overall accuracies to predict CF% in anterior and posterior were 66.6 and 73.7 %, respectively. For the anterior group, the accuracies in each category were (shown in Figure 3.4) 94.3, 40.5, 33.5, 32.7, and 46.2 % for CF1, CF2, CF3, CF4, and CF5, respectively. For the posterior group, the accuracies in each category were (shown in Figure 3.5) 86.9, 49.6, 35.1, 23.5, and 37.5 % for CF1, CF2, CF3, CF4, and CF5, respectively. When comparing anterior group to posterior group, SMS had better overall accuracy in the posterior group.

The overall accuracy for RIMF to predict CF% is 60.8 %. Accuracies for RIMF were 82.6, 41.4, 15.4, 11.6, and 5.9 % when predicting CF1, CF2, CF3, CF4, and CF5, respectively (shown in Figure 3.6). RIMF had the best accuracy in predicting CF% under 2, whereas when predicting CF% at a higher percentage, the accuracy lowered. When looking at the distribution of

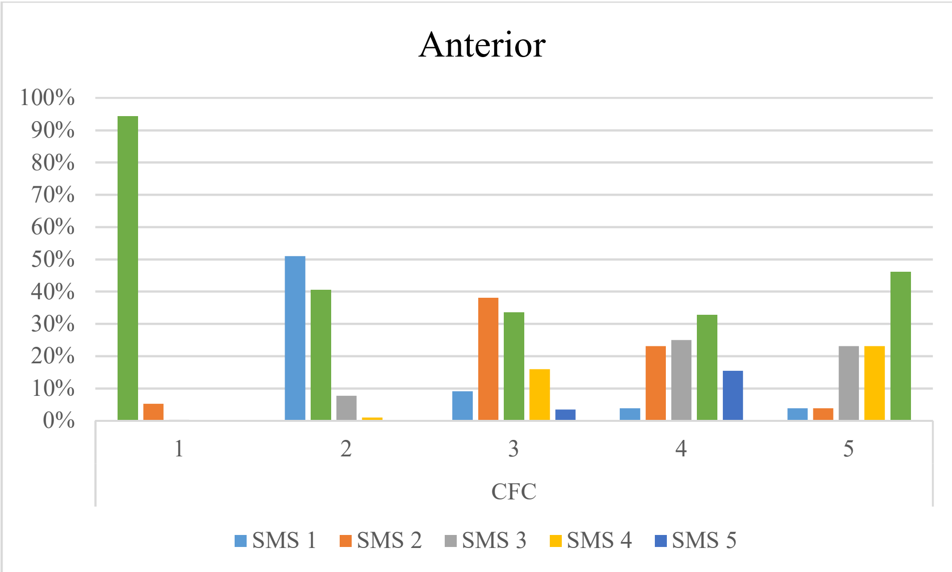


Figure 3.4. Anterior crude fat categorical versus Subjective. Within a crude fat category, the distribution is what the samples were called subjectively and the percentages will sum to 100. The green column represents the accuracy of calling subjective marbling score in each category, whereas light blue, orange, gray, yellow, and dark blue is what the samples were called subjectively.

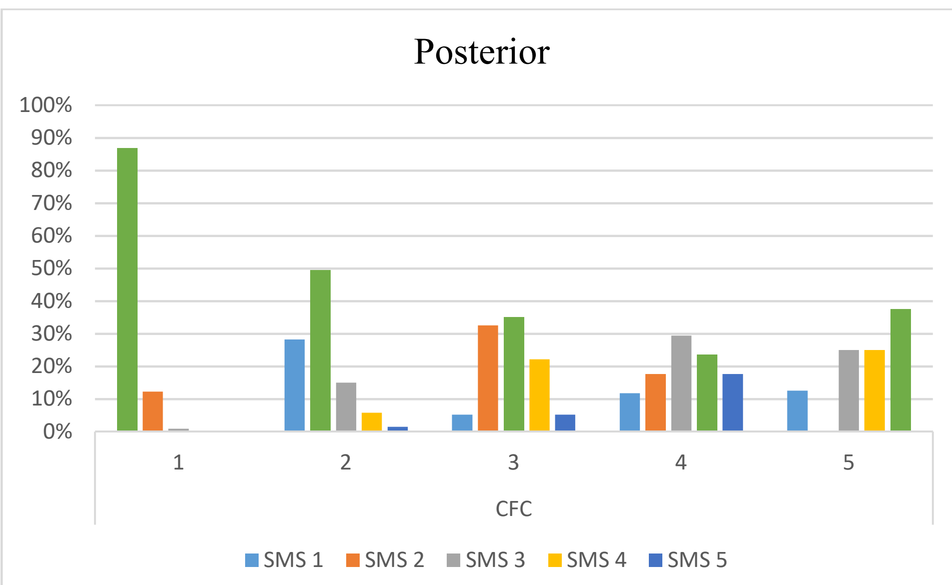


Figure 3.5. Posterior crude fat categorical versus Subjective. Within a crude fat category, the distribution is what the samples were called subjectively and the percentages will sum to 100. The green column represents the accuracy of calling subjective marbling score in each category, whereas light blue, orange, gray, yellow, and dark blue is what the samples were called subjectively.

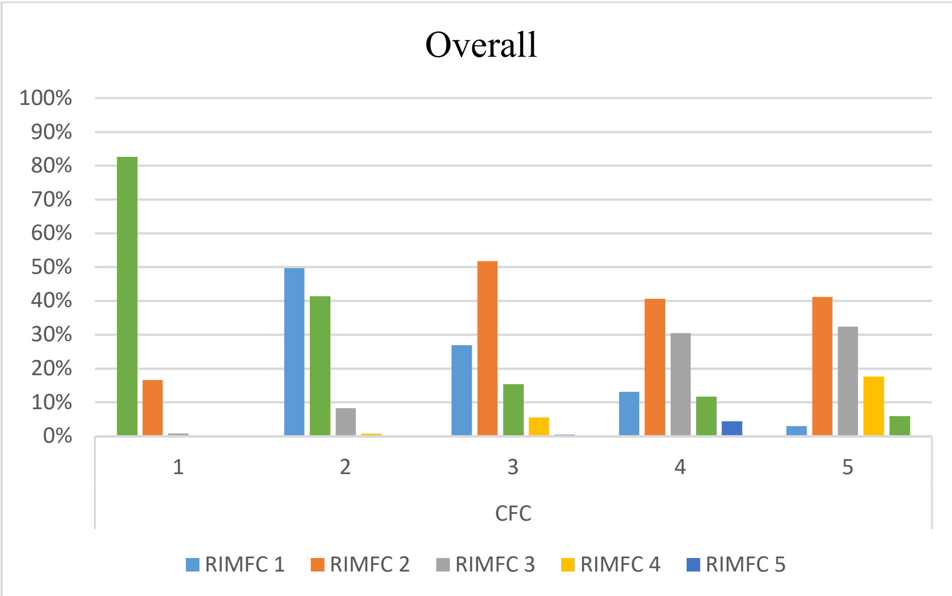


Figure 3.6. Overall crude fat categorical versus RIMFC. Within a crude fat category, the distribution is what the samples were called subjectively and the percentages will sum to 100. The green column represents the accuracy of calling subjective marbling score in each category, whereas light blue, orange, gray, yellow, and dark blue is what the samples were assessed.

prediction of CF%, RIMF results in predicting the CF% lower than the actual CF%. This could be due to the insufficient of sample size in high CF% as the majority of the samples were CF1 and CF2. When dividing the pork chop into anterior (3rd rib) and posterior (10th rib), similar results were found. The overall accuracies to predict CF% in anterior and posterior were 56.6 and 71.1%, respectively. For the anterior group, the accuracies in each category were 74.9, 48.9, 19.9, 19.2, and 11.5 % for CF1, CF2, CF3, CF4, and CF5, respectively (shown in Figure 3.7). For the posterior group, the accuracies in each category were 88.3, 41.2, 14.3, 11.8, and 0.0 % for CF1, CF2, CF3, CF4, and CF5, respectively (shown in Figure 3.8). When comparing anterior group to posterior group, RIMF showed better predictive power in the posterior group.

When comparing SMS and RIMF, both had the best accuracies at lower CF%. However, when predicting higher CF% (4 or 5), SMS had better accuracies as well as a narrower distribution of the prediction when compared to RIMF. Both SMS and RIMF had higher accuracy when predicting CF% in the posterior chop when compared to the anterior chop. It is

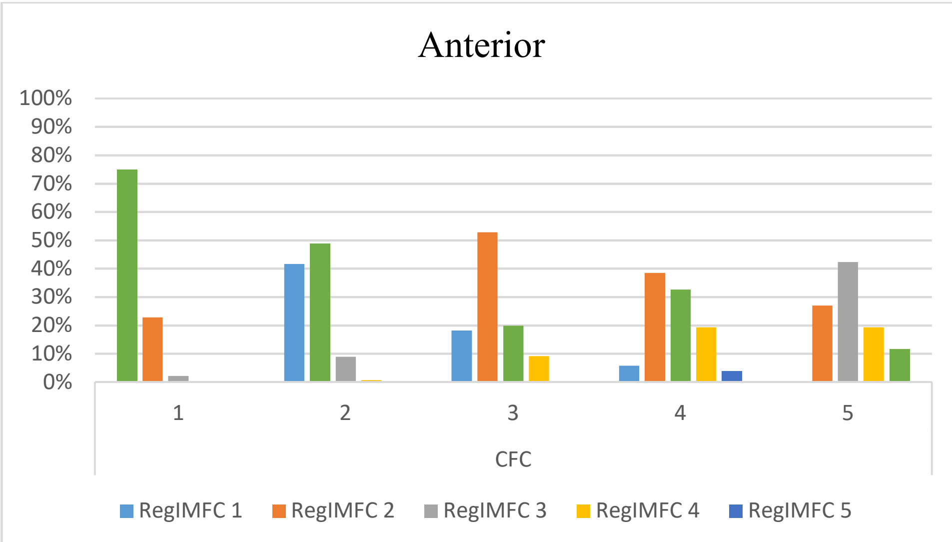


Figure 3.7. Anterior crude fat categorical versus RIMFC. Within a crude fat category, the distribution is what the samples were called subjectively and the percentages will sum to 100. The green column represents the accuracy of calling subjective marbling score in each category, whereas light blue, orange, gray, yellow, and dark blue is what the samples were assessed.

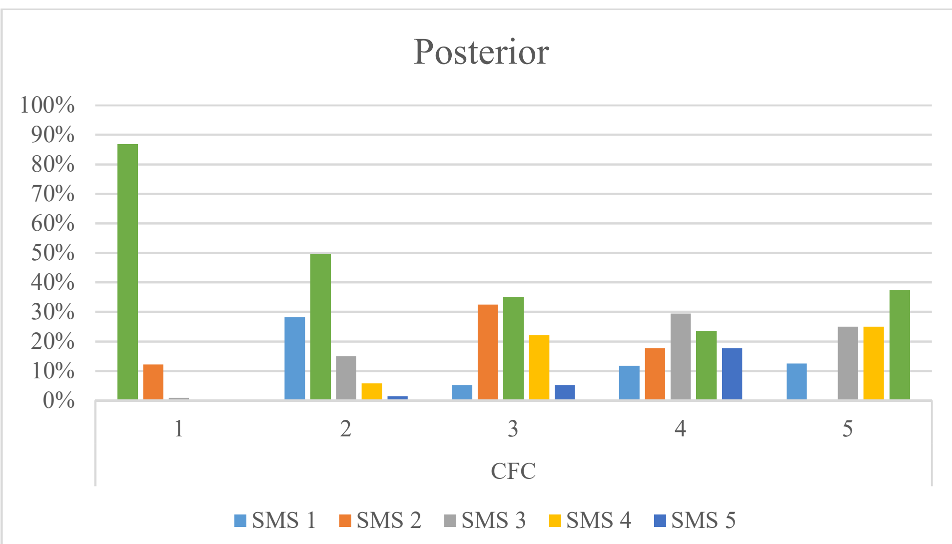


Figure 3.8 Posterior crude fat categorical versus RIMFC. Within a crude fat category, the distribution is what the samples were called subjectively and the percentages will sum to 100. The green column represents the accuracy of calling subjective marbling score in each category, whereas light blue, orange, gray, yellow, and dark blue is what the samples were called.

noticed that when comparing anterior to posterior chops, anterior chops tended to be lighter in color (data not presented) and have higher CF% than posterior chops. Thus, the combination of paler lean meat and an increase in CF% increased the difficulty to distinguish between IMF and lean meat, especially for CVS.

Residual Distributions

Residual distributions overall and by plant are shown for IIMF in Fig. 3.9 and for RIMF in Fig. 3.10. Overall accuracies (residual between -0.5 and +0.5) were 20.9 and 53.3 % for IIMF and RIMF, respectively. For the individual plants, the accuracies were 31.5, 28.0, 27.5, 27.3, 14.8, 8.8, and 8.8 % for IIMF and 46.8, 42.2, 51.6, 56.3, 57.9, 57.6, and 60.9 % for RIMF for plants 1, 2, 3, 4, 5, 6, and 7, respectively. The residual distribution was right-skewed for IIMF, suggesting that CVS tends to underestimate CF% when using just IIMF. However, when switching to RIMF, the residual distribution shifts towards being more accurate. However, there is a slight overestimation (left skewness) of CF%. This could possibly be improved by ensuring that the regression model development data set has equal representation of samples in each CF% category.

Residual distributions of anterior chops overall and by plant are shown for IIMF in Fig. 3.11 and for RIMF in Fig. 3.12. Overall accuracies (residual between -0.5 and +0.5) were 16.2 and 47.3 % for IIMF and RIMF, respectively. For the individual plants, the accuracies were 22.0, 23.5, 20.0, 19.8, 15.0, 8.5, and 5.5 % for IIMF and 44.5, 39.5, 44.0, 54.5, 49.5, 45.5, and 55.0 % for RIMF for plants 1, 2, 3, 4, 5, 6, and 7, respectively. The residual distribution was right-skewed for IIMF, suggesting that CVS tends to underestimate CF% when using just IIMF. The

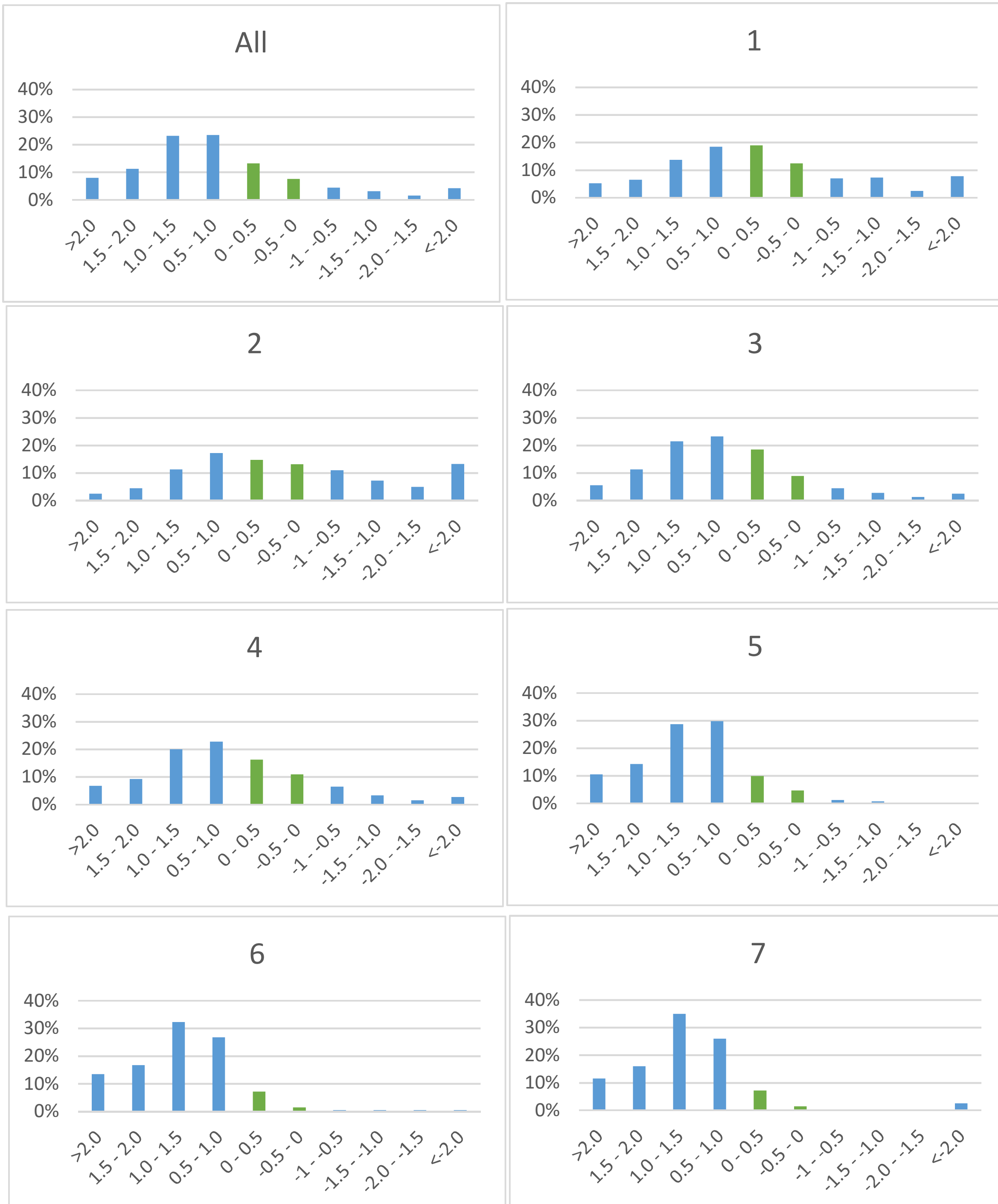


Figure 3.9. Overall residual distribution of image intramuscular fat percentage versus crude fat percentage. Green column represents percentage of residual that were within -0.5 and + 0.5.

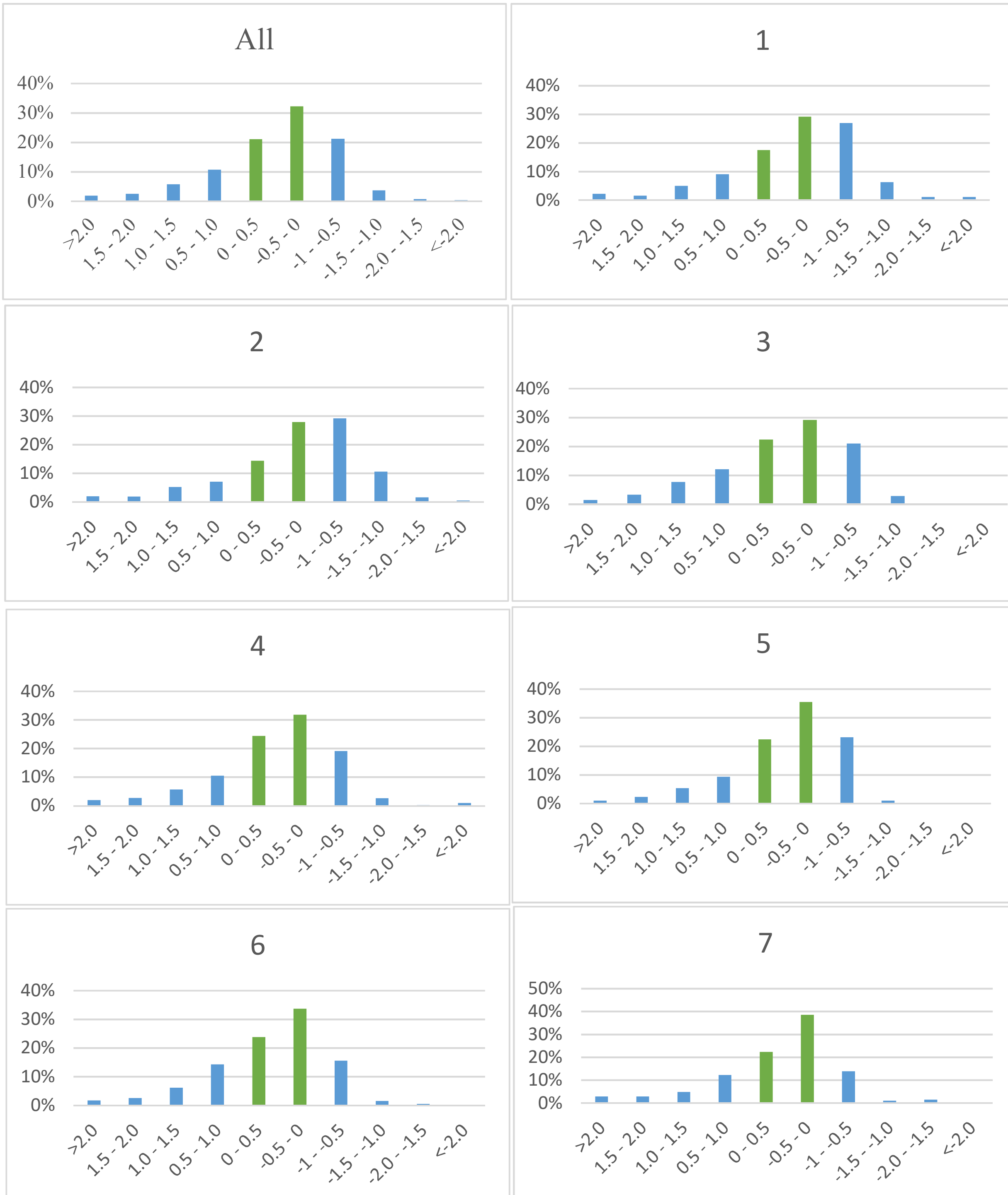


Figure 3.10. Overall residual distribution of regression intramuscular fat percentage versus crude fat percentage. Green column represents percentage of residual that were within -0.5 and + 0.5.

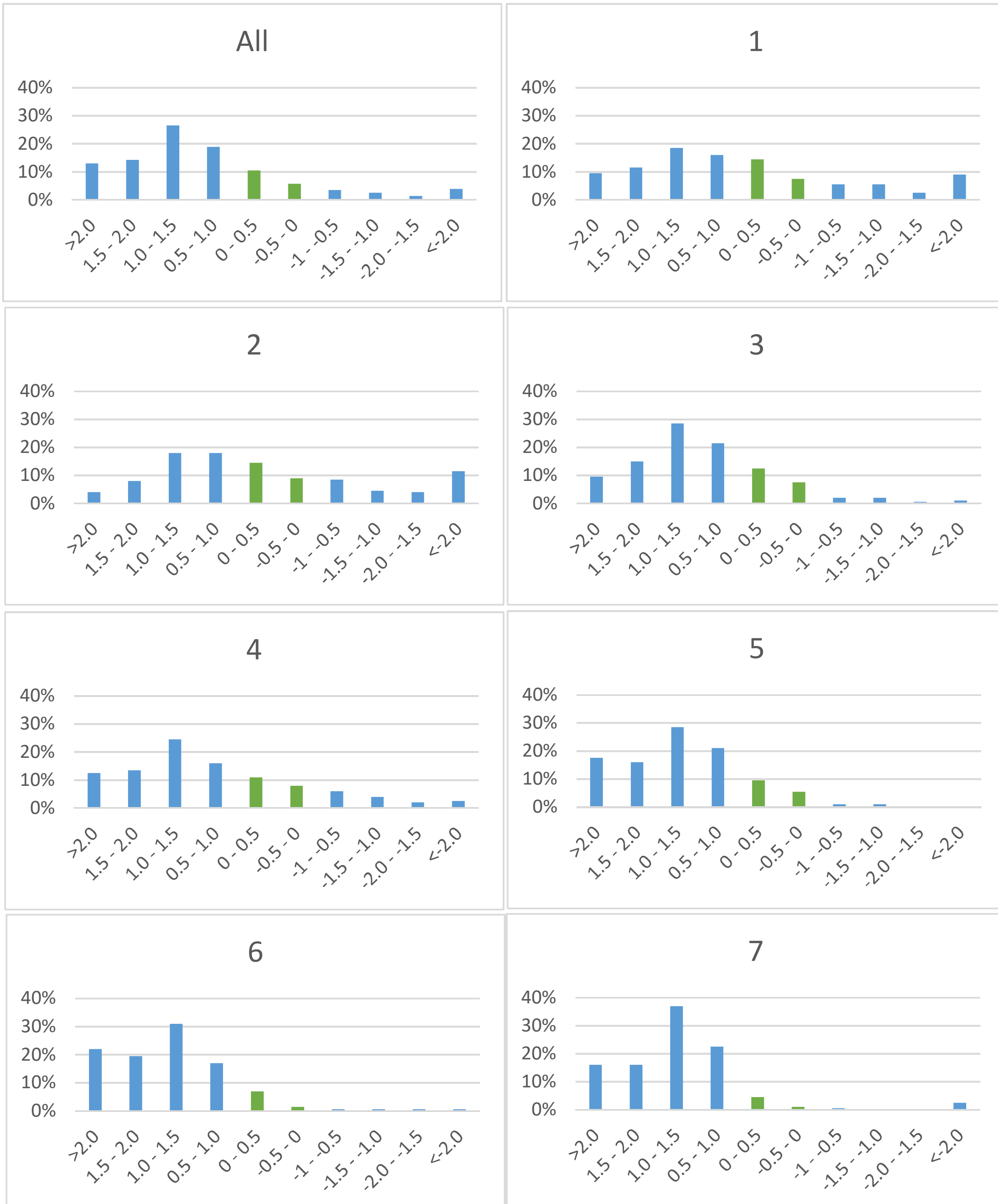


Figure 3.11. Anterior residual distribution of image intramuscular fat percentage versus crude fat percentage. Green column represents percentage of residual that were within -0.5 and + 0.5.

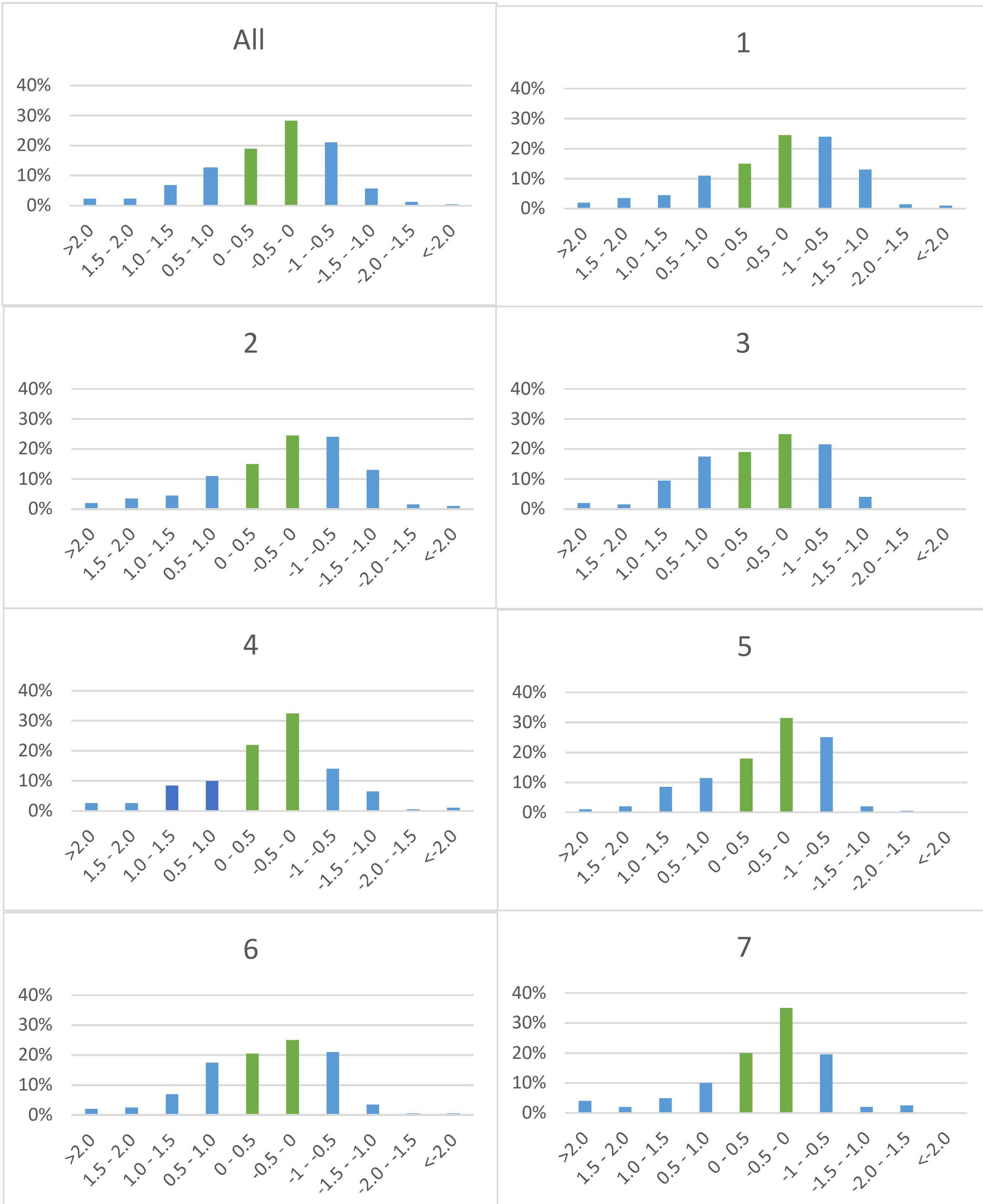


Figure 3.12. Anterior residual distribution of regression intramuscular fat percentage versus crude fat percentage. Green column represents percentage of residual that were within -0.5 and + 0.5.

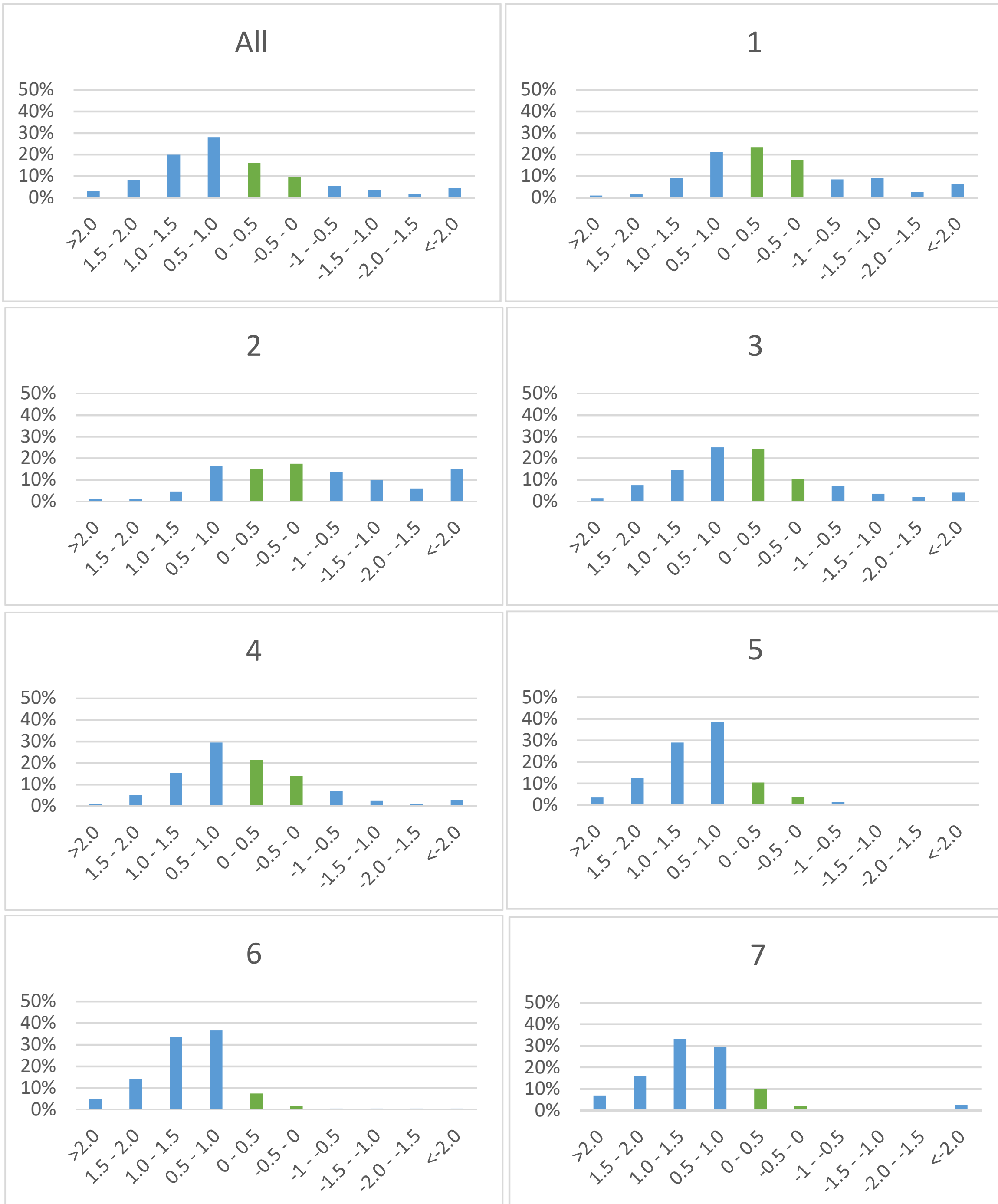


Figure 3.13. Posterior residual distribution of image intramuscular fat percentage versus crude fat percentage. Green column represents percentage of residual that were within -0.5 and + 0.5.

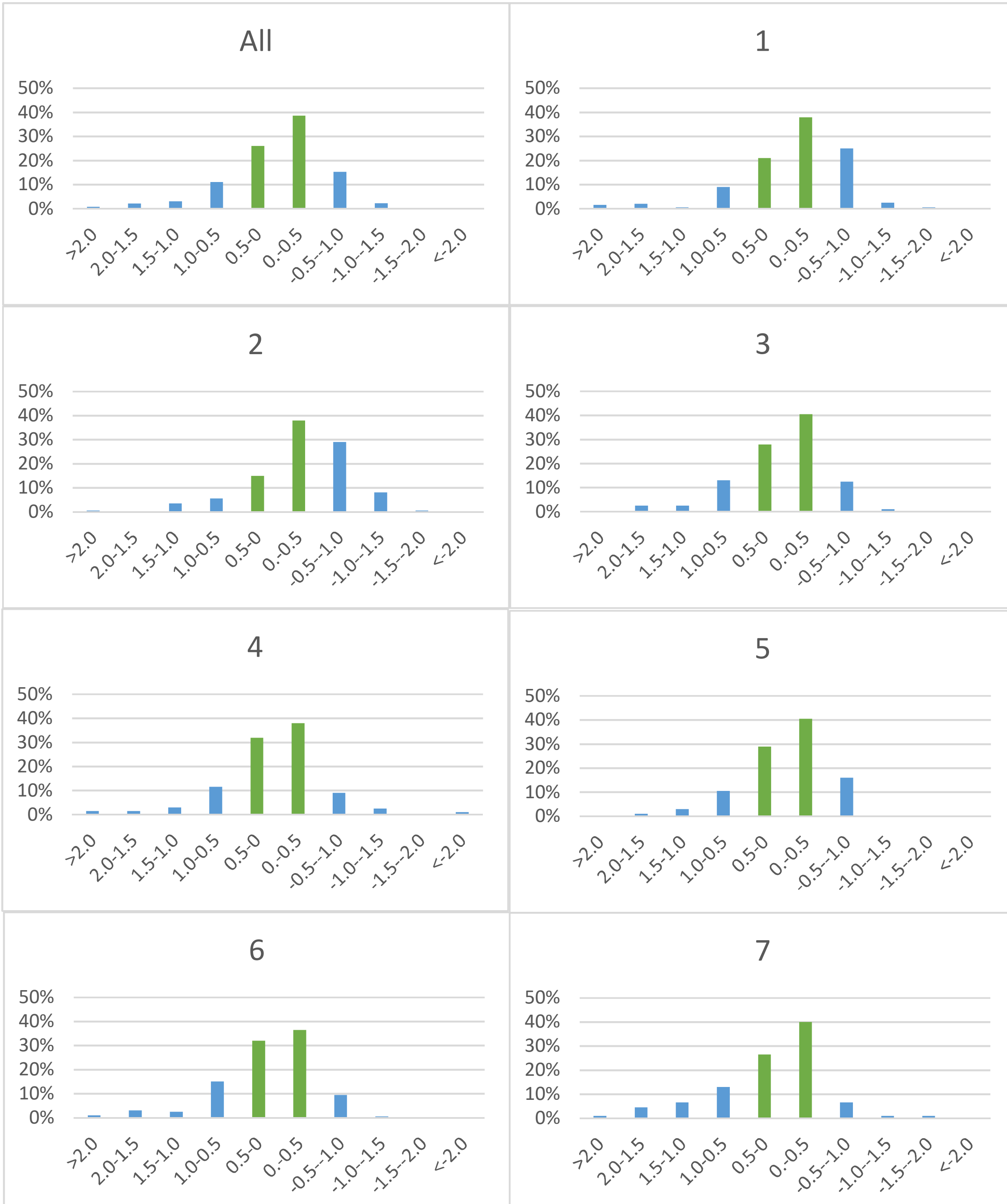


Figure 3.14. Posterior residual distribution of regression intramuscular fat percentage versus crude fat percentage. Green column represents percentage of residual that were within -0.5 and + 0.5.

distribution of residuals were wider when compared to overall and the accuracies dropped. This showed similarity with SMS in assessment of anterior chop. However, when switching to RIMF, the residual distribution shifts towards being more accurate. However, there is a slight overestimation (left skewness) of CF%. This could possibly be improved by ensuring that the regression model development data set has equal representation of samples in each CF% category.

Residual distributions of posterior chops overall and by plant are shown for IIMF in Fig. 3.13 and for RIMF in Fig. 3.14. Overall accuracies (residual between -0.5 and +0.5) were 25.7 and 64.7 % for IIMF and RIMF, respectively. For the individual plants, the accuracies were 41.0, 32.5, 35.0, 35.5, 14.5, 9.0, and 10.0 % for IIMF and 59.0, 53.0, 68.5, 70.0, 69.5, 68.5, and 66.5 % for RIMF for plants 1, 2, 3, 4, 5, 6, and 7, respectively. The residual distribution was right-skewed for IIMF, suggesting that CVS tends to underestimate CF% when using just IIMF. The distribution of residuals were narrower when compared to overall and the accuracies increased. This showed similarity with SMS in assessment of anterior chop. However, when switching to RIMF, the residual distribution shifts towards being more accurate. However, there is a slight overestimation (left skewness) of CF%. When comparing results of anterior chops and posterior chops, both SMS and CVS has shown better accuracies in predicting CF% of 10th rib chop, this suggest that there are discrepancy in between anatomy site.

Conclusion

When comparing results for prediction of CF%, SMS had an overall higher accuracy (70.1 %) when compared to RIMF (62.3 %). When dividing the pork chops into anterior and posterior groups, both SMS (66.6 %) and RIMF (56.6 %) had lower accuracies for anterior chops and both SMS (73.7 %) and RIMF (71.1 %) had higher accuracies for posterior chops. When using IIMF as a predictor for CF%, the overall accuracy was 36.8 % and the distribution of residuals was right skewed, suggesting that the CVS tends to underestimate CF%. When using RIMF% as predictor for CF%, the overall accuracy increased to 53.3 % and the distribution of residual was only slightly left skewed, suggesting a slight tendency to overestimate CF%. Overall, the results demonstrate the potential of using CVS as an objective predictor of CF% in pork chops.

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CHAPTER 4. PREDICTING PORK LOIN INTRAMUSCULAR FAT USING VISION SYSTEM TECHNOLOGY

Abstract

The objective of this study was to examine the potential of using computer vision system (CVS) as a tool to predict pork loin intramuscular fat (IMF) percentage under industry scale equipment and environment. Whole pork loins (n=1400) were obtained from 7 major meat plants. Color images of pork loin chop samples (n=1040) were acquired using a computer vision system. Subjective marbling scores (NPB, 2011) were determined by a trained evaluator according to surface of loin. Crude fat percentage (CF%) of 3rd and 10th rib chops were calculated using ether extract method (AOAC, 1990) and averaged to represent CF% of the whole loin. Results show that subjective marbling scores had an overall accuracy of 53.3 % of predicting CF%, while for individual categories, the accuracies were 66.2, 39.9, 31.3, 17.4, and 60.0% for CF% ranges of 0-1.99, 2.00-2.99, 3.0-3.99, 4.0-4.99, and 5.0-5.99 %, respectively. The overall accuracy of using CVS was 58.56 %, while for individual catergories, the accuracies were 75.1, 46.0, 21.9, 8.7, and 0 % for CF% ranges of 0-1.99, 2.00-2.99, 3.0-3.99, 4.0-4.99, and 5.0-5.99 %, respectively. These results indicate that CVS is better at predicting CF% in pork whole loins when compared to subjective marbling score and have the potential of replacing subjective marbling scores.

Introduction

Marbling is an important factor which positively influences meat quality. In the beef industry, quality is primarily driven by the level of marbling. In the pork industry, marbling has been shown to be associated with greater juiciness, flavor, and tenderness scores in center-cut loin chops (Fernandez et al., 1999b; Brewer et al., 2001; Cannata et al., 2010; Moeller et al., 2010a,b), with pork containing more intramuscular fat (IMF) being more favorable than pork containing low amounts of IMF (Brewer et al., 2001; Font-i-Furnols et al., 2012).

While the preference in cooked pork is for greater IMF, visual acceptability levels of IMF in uncooked pork differs between countries. Well-marbled pork, or pork containing high levels of IMF, are preferred in Asian countries such as Japan and Korea while lean pork, or pork containing low levels of IMF, are preferred in European countries such as Finland and Poland (Ngapo et al., 2007). In the US, the amount of visible fat in loin chops was found to have a negative effect on overall appearance acceptability to consumers (Brewer et al., 2001). This shows variety in the acceptability of IMF in pork based on cultural background.

Newman (2015) showed that a great deal of pork quality variation exists in the retail meat case nationwide and that the majority of pork (over 80 %) has a IMF percentage (IMF%) which lies within 1 to 3 %. Currently there are limited methods to measure IMF% in pork. In laboratory-based research, IMF% can be determined by using crude fat extraction (AOAC, 1990); however, this extraction method is labor-intensive, time-consuming, and uses up the actual samples. In pork processing plants, marbling is graded subjectively by trained evaluators on a scale of 1 to 10 (1 = devoid of marbling, 10 = abundant of marbling; NPB, 2011). However, subjective marbling is inconsistent due to the different evaluators and environmental factors in the plant such as lighting condition. An objective, or instrumental, measurement of IMF%, similar to using a colorimeter for determining color, would benefit the pork industry by providing a quick and effective means for measuring quality or sorting loins for different market specifications. Such an instrument could also be helpful in providing a more consistent and high quality product to consumers.

Computer vision systems (CVS) contain an illumination system, a camera, and image analyzing software and have been widely utilized in the food industry as an inspection tool. It has great features such as being rapid, economic, consistent, accurate, and non-invasive (Sun, 2000). In the food industry, it has been widely used for different features measurement such as detection or grading to differentiate color, size, texture features, shape, and uniformity of the product (Sun, 2000). More pertinent to this research, CVS has been utilized in the beef industry to objectively measure multiple features of beef quality such as marbling and yield percentage using the "beef cam" (Cannell et al., 2002). Research has shown the potential for CVS in analyzing, or predicting, beef color (Larraín et al., 2008), fat color, (Chen et al., 2010), tenderness (Li et al., 1999, 2001; Tan, 2004; ElMasry et al., 2012; Sun et al., 2012), pH value (ElMasry et al., 2012), and marbling (Chen et al., 2010; Jackman et al., 2009). More recent research has been done evaluating the potential use of CVS in the pork industry, with research focusing on classification or detection of pale, soft, and exudative pork (Warriss et al, 2011; Chmiel et al, 2016), pork color grading (Sun et al., 2016), IMF% (Lu et al., 2000; Faucitano, et al., 2005; Huang et al., 2013; Liu & Ngadi, 2014; Sun et al., 2016), and *Escherichia coli* contamination (Tao & Peng, 2014).

While using the 10^{th} rib pork chop as a sample source is common in research, industry uses the whole loin to sort to different markets rather than cutting the loin at the 10^{th} rib, which sabotages the integrity of the product and devalues the product. Therefore, the objective of this study was to evaluate the effectiveness of using an industrial CVS as a consistent, accurate, and non-invasive method to estimate IMF% of whole, boneless pork loins.

Materials and Methods

Sample Collection and Image Acquisition

Whole, boneless loins were obtained from seven different processing plants (n = 200 per plant). Each sample was selected by a trained evaluator from the deboning line. Samples were chosen to maximize the variation in pork quality for subjective color (SCS) and marbling (SMS) scores, which were assessed on-line according to National Pork Board (NPB) standards (NPB, 2011). After loins were selected and removed from the deboning line, an image of the lean surface of the loin was acquired using a CVS (Fig. 4.1), consisting of an industry camera (NI 1776C smart camera, National Instrument, Ltd., USA) with a 1/1.8" F1.6/4.4-11-mm lens (LMVZ4411, Kowa, Ltd., Japan), a 44-inch dome light (DL180, advance illumination, Ltd., USA), and a personal laptop (Lenovo, Ltd., China). The CVS was attached to a table to ease

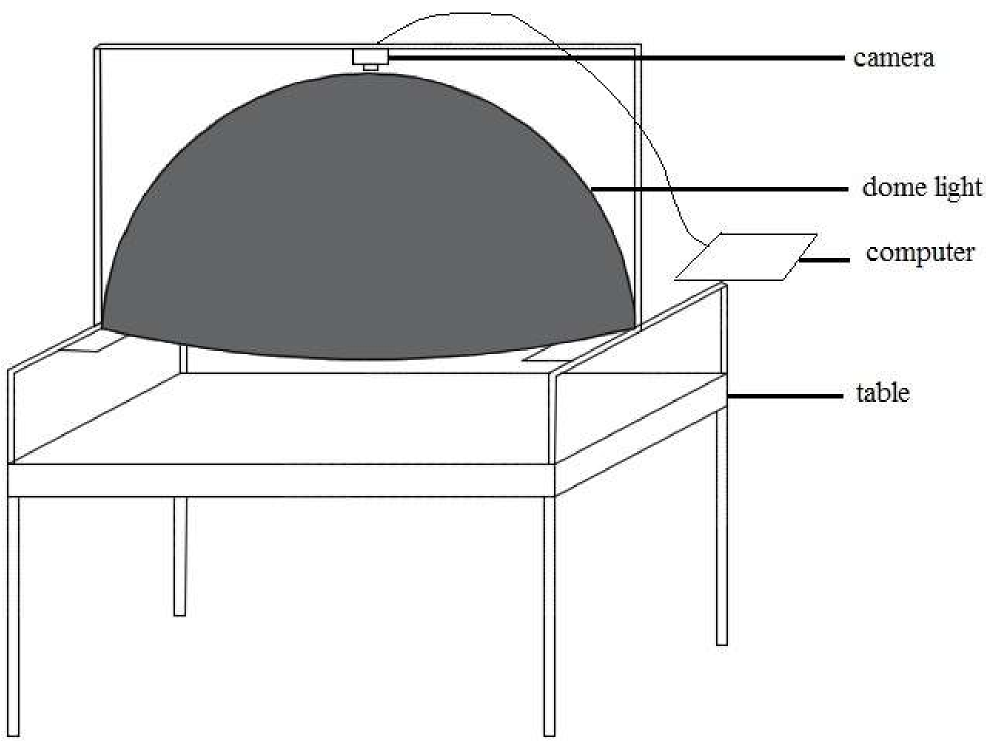

Figure 4.1. Computer vision system used for this project and it's key component including the camera, dome light, and computer.

transportation of the dome light and to standardize the relationship of the camera to the dome light and the samples. A black, light-absorbent fabric was installed between the dome light and table to exclude light noise from the surrounding environment. Before each plant collection, a Minolta white tile was used for calibration. The white tile was placed in the center and corner of the CVS to ensure the evenness of light spread. When taking pictures of the white tile, color space red green blue color features were extract and used as standards for calibration and setting of the CVS. Each sample was manually placed on a light-absorbing, black background surface for image acquisition. The color image was captured and stored using LabVIEW software (National instrument, Ltd, TX).

After images were acquired, pork loins were vacuum packaged and transported in a refrigerated truck to the US Meat Animal Research Center in Clay Center, NE. Loins were stored at 4 °C for 14 d. After 14 d, whole loins were cut into 2.54 cm thick chops. The 3rd and 10th rib chops were collected, vacuumed packaged, and transported to North Dakota State University to determine crude fat percentage (CF%). After arrival at North Dakota State University, chops were trimmed of connective tissue and subcutaneous fat and then freeze-dried for 48 h to remove moisture. After the freeze-drying period, CF&&% was determined gravimetrically using Soxhlet extraction with petroleum ether according to AOAC procedure (AOAC, 1990). The average of the 3rd and 10th rib chops were used to represent the CF% of the entire loin.

Image Analysis

An original pork sample image acquired by the CVS is shown in Fig. 4.2(a). To remove the background of the image automatically, Otsu method was adopted and performed using the LabVIEW software (Fig. 4.2(b); Otsu, 1975). Once the background is removed the region of interest (ROI; 5.08 × 9.18 cm) was then determined automatically using a mapping system to avoid uncleaned surface or connective tissue remained on the pork loin surface (Fig. 4.2(c)). After determination of ROI, the Sobel Method was applied to the image to segment lean muscle pixels and IMF pixels (Fig. 4.2(d)). After segmentation, the pixels were then counted to calculate image IMF% (IIMF) within ROI (Fig. 4.2(e)).

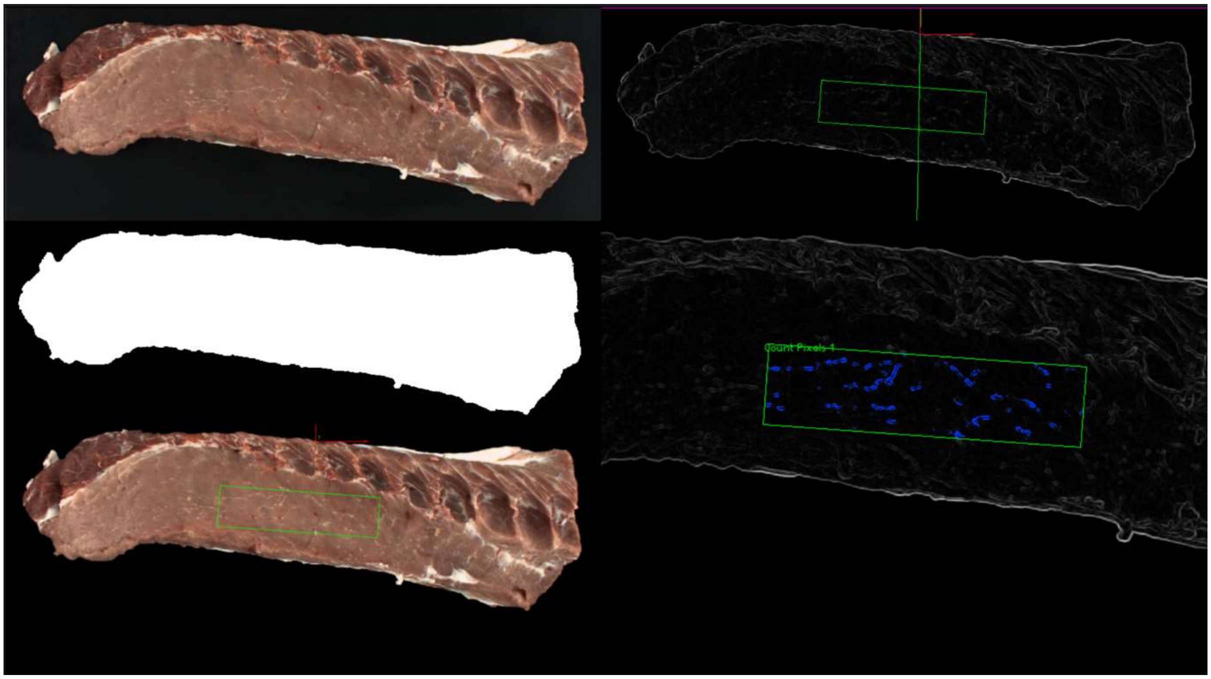

Figure 4.2. Segmentation procedure: (a) Original pork sample image. (b) Removal of background. (c) Selection of region of interest (d) Applying Sobel detection. (e) Calculation of intramuscular fat pixel percentage within the region of interest.

Data Analysis

In order to calculate the accuracies of SMS predicting CF%, CF% was categorized from range of 0-1.99, 2.00-2.99, 3.00-3.99, 4.00-4.99, and 5.00 % and above as CF1, CF2, CF3, CF4, and CF5, respectively. The bootstrap method by Efron (1979) was adopted using SAS (v. 9.4; SAS Institute, Inc., Cary, NC). Data were set to randomly divide data base into training group 70% and test group 30% for 100 replications. The 100 simple regression equations were then averaged to generate a final regression equation to calculate regression IMF% (RIMF) using the

IIMF. To calculate the accuracies of CVS predicting CF%, RIMF was categorized into RIMF1, RIMF2, RIMF3, RIMF4 and RIMF5 using the same percentages as CF%. The residual was also calculated as CF% minus RIMF to further understand the prediction of RIMF. All procedures were accomplished using SAS

Results and Discussion

Distribution of CFC in Pork Loin

From 1045 loins that were collected the distribution of CF% were 785, 441, 129, 31, and 14 respectively in CF1, CF2, CF3, CF4, and CF5. Which by the definition of SMS if graded correctly there should be 56.1%, 31.5%, 9.2%, 2.2%, and 1.0% that were graded respectively SM1, SM2, SM3, SM4, and SM5. This suggest that the majority of CF% distribution of pork loins were below 3% (87.59%).

Accuracies

When comparing CF% with SMS, the overall prediction is 53.30%. When evaluating different levels of marbling, SMS had an accuracy of 66.2, 40.0, 31.2, 17.4, and 60.0 % when predicting CF1, CF2, CF3, CF4, and CF5, respectively (shown in Fig. 4.3). SMS had the best accuracy in predicting CF% under 2, whereas when predicting CF% at a higher percentage the accuracies were lowered. In fact, Fig. 4.3 shows that the spread is wide but not skewed. This could be explained by several factors. Variety in pork appearance can affect SMS. While the NPB standards provide an example for each SMS that corresponds to the CF% of pork chop, however, when grading on whole pork loin, there are different traits that affect the evaluator's ability to correctly assess the CF%. Unlike pork chop, pork loin may display a sizable variety in color or marbling from posterior end to anterior end, therefore, increasing the difficulty for evaluator to accurately assess pork quality. Additionally, assessment of whole loin may be interfered by the different standard of trimming from plant to plant, as more connective tissue remains, less lean meat surface is revealed. this suggest that even for trained grader it is hard to subjectively predict the marbling of loin by observing the surface of loins when CF% are within the 2-5% range.

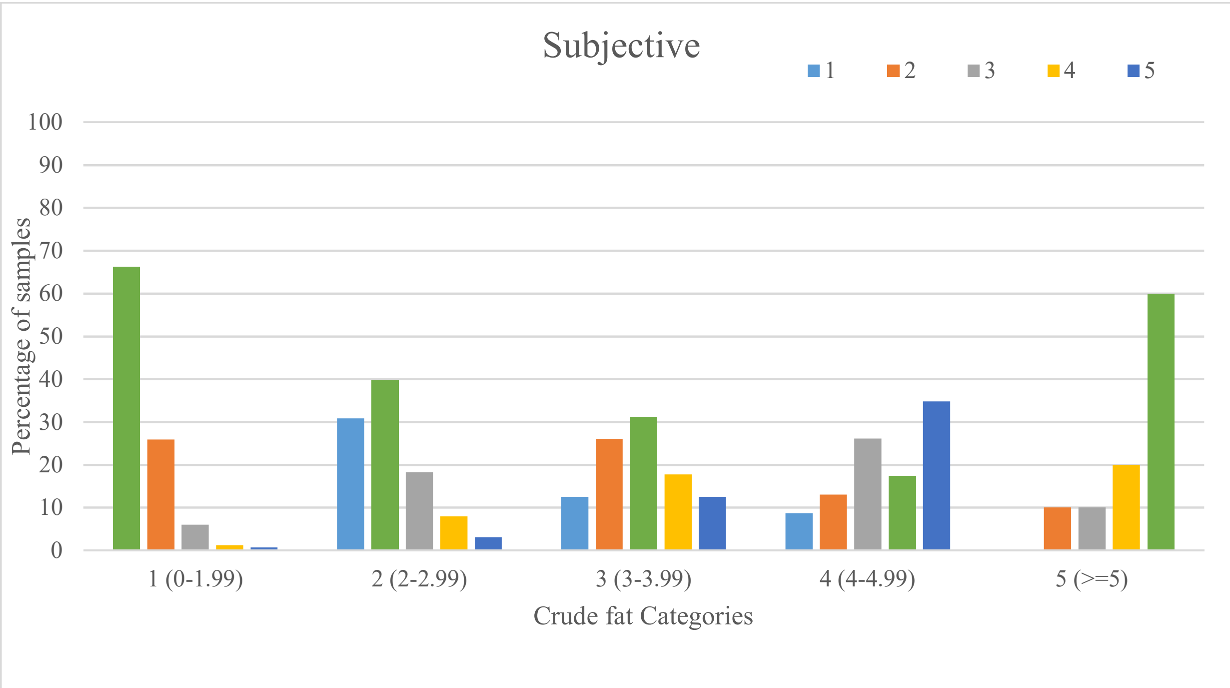

Figure 4.3. Crude fat categorical versus subjective marbling scores. Within a crude fat category, the distribution is what the samples were called subjectively and the percentages will sum to 100. The green column represents the accuracy of calling subjective marbling score in each category.

The overall accuracy for RIMF% to predict CF% was 58.56%. Accuracies for RIMF were 75.1, 46.0, 21.9, 4.3, and 0.0 % when predicting CF1, CF2, CF3, CF4, and CF5, respectively (shown in Fig. 4.4). RIMF had the best accuracy in predicting CF% under 2, whereas when predicting CF% at a higher percentage, the accuracy lowered. When looking at the distribution of prediction of CF%, RIMF predicted the CF% lower than the actual CF%. This could be due to the insufficient of sample size in high CF% as the majority of the samples were CF1 and CF2.

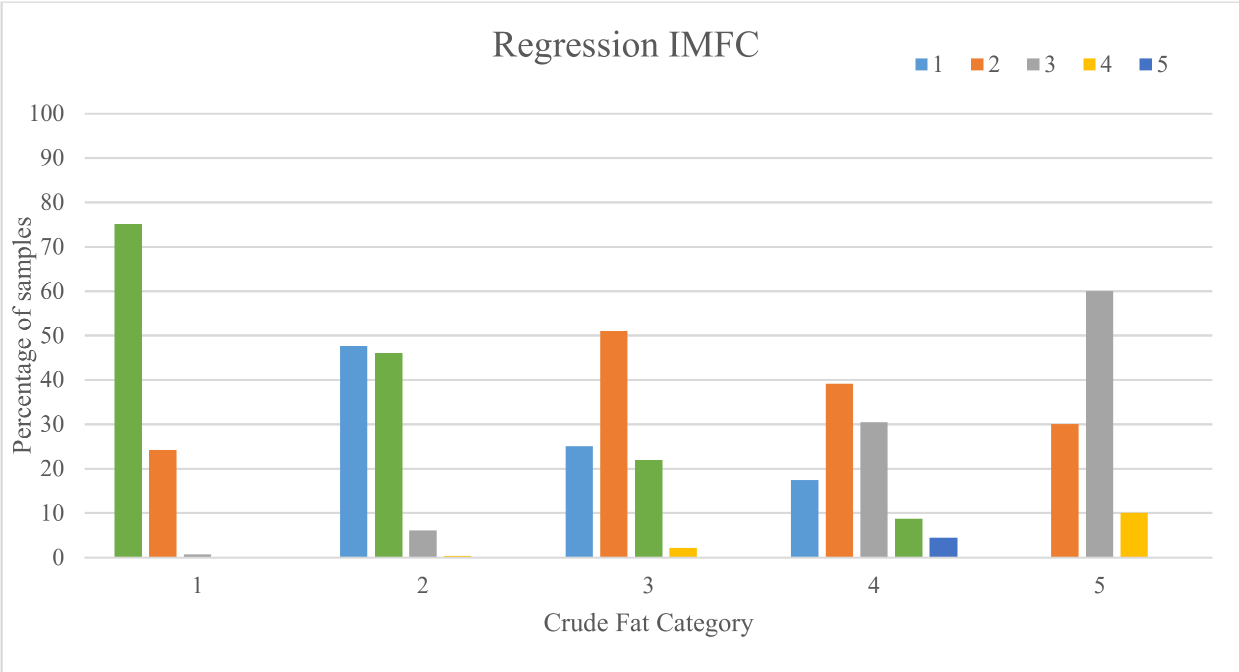

Figure 4.4. Crude fat categorical versus regression intramuscular fat categorical. Within a crude fat category, the distribution is what the samples were estimated as using the regression equation and the image intramuscular fat estimates and the percentage will sum up to 100. The green columns represent the correct prediction within each category.

When comparing crude fat percentage prediction accuracies between RIMF and SMS, RIMF has shown a higher overall accuracy (58.5% vs 53.3%). However, in each category, RIMF only had a higher accuracy in category 1, this result could be due to the imbalance amount of sample within each category, while SMS has shown 60.0% in category 5, it only represents 1% of the sample.

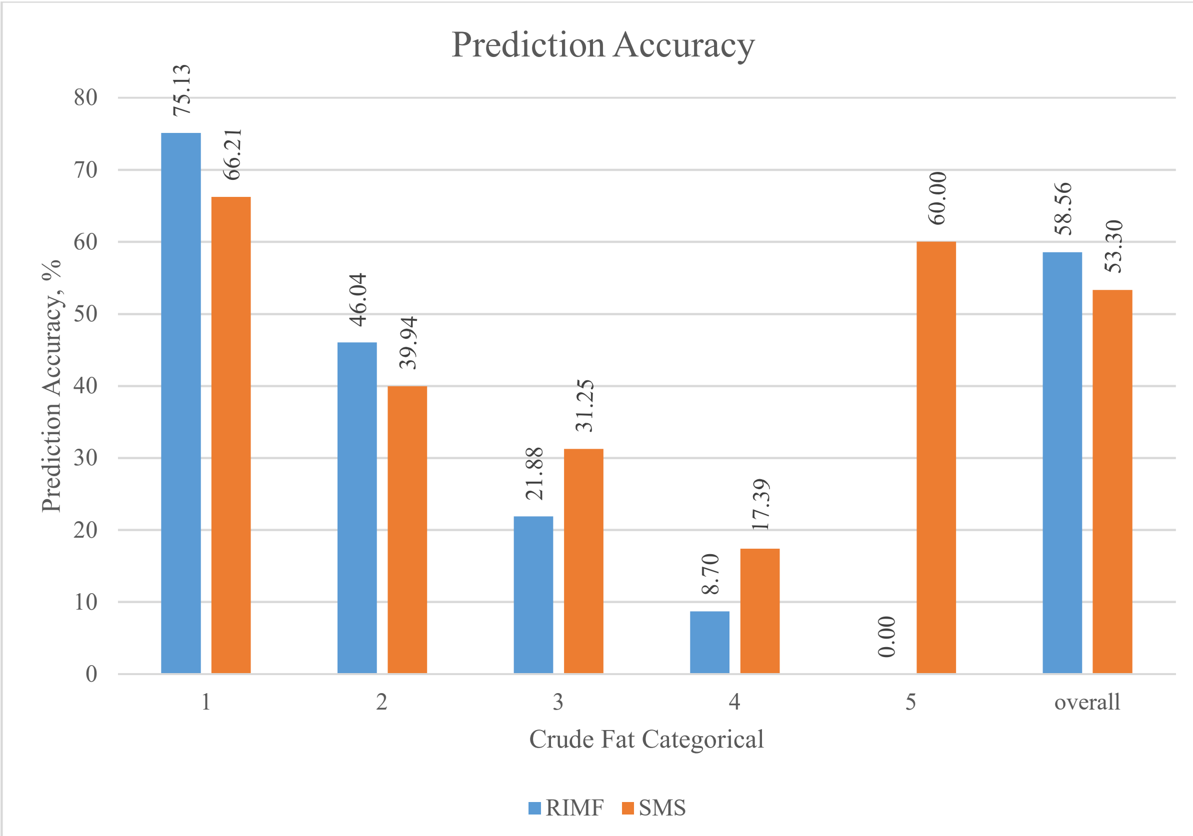

Figure 4.5. Accuracies for regression intramuscular fat categorical (blue bars) and subjective marbling scores (orange bars) for predicting crude fat categorical by category and overall.

Residual Distribution

Residual distributions overall and by plant are shown for IIMF in Fig. 4.6 and for RIMF in Fig. 4.7 Overall accuracies (residual between -0.5 and +0.5) were 24.5 and 53.3 % for IIMF and RIMF, respectively. For the individual plants, the accuracies were 31.7, 2.8, 30.1, 31.5, 14.9, 42.6, and 39.5 % for IIMF and 33.8, 49.2, 48.6, 60.4, 59.6, 62.8, and 49.0 % for RIMF for plants 1, 2, 3, 4, 5, 6, and 7, respectively. A severe of inconsistent of residual distribution was observed for IIMF, in plant 1, 2, 4, 6, and 7 the distribution of the residuals were left skewed while in plant 3 and 5 the distribution of residuals were right skewed. There wasn't a tendency to overestimate or underestimate when using IIMF% to predict CF% of pork loin, but rather both. This could be due to the following reasons: 1. The possibility of calculating connective tissue or subcutaneous

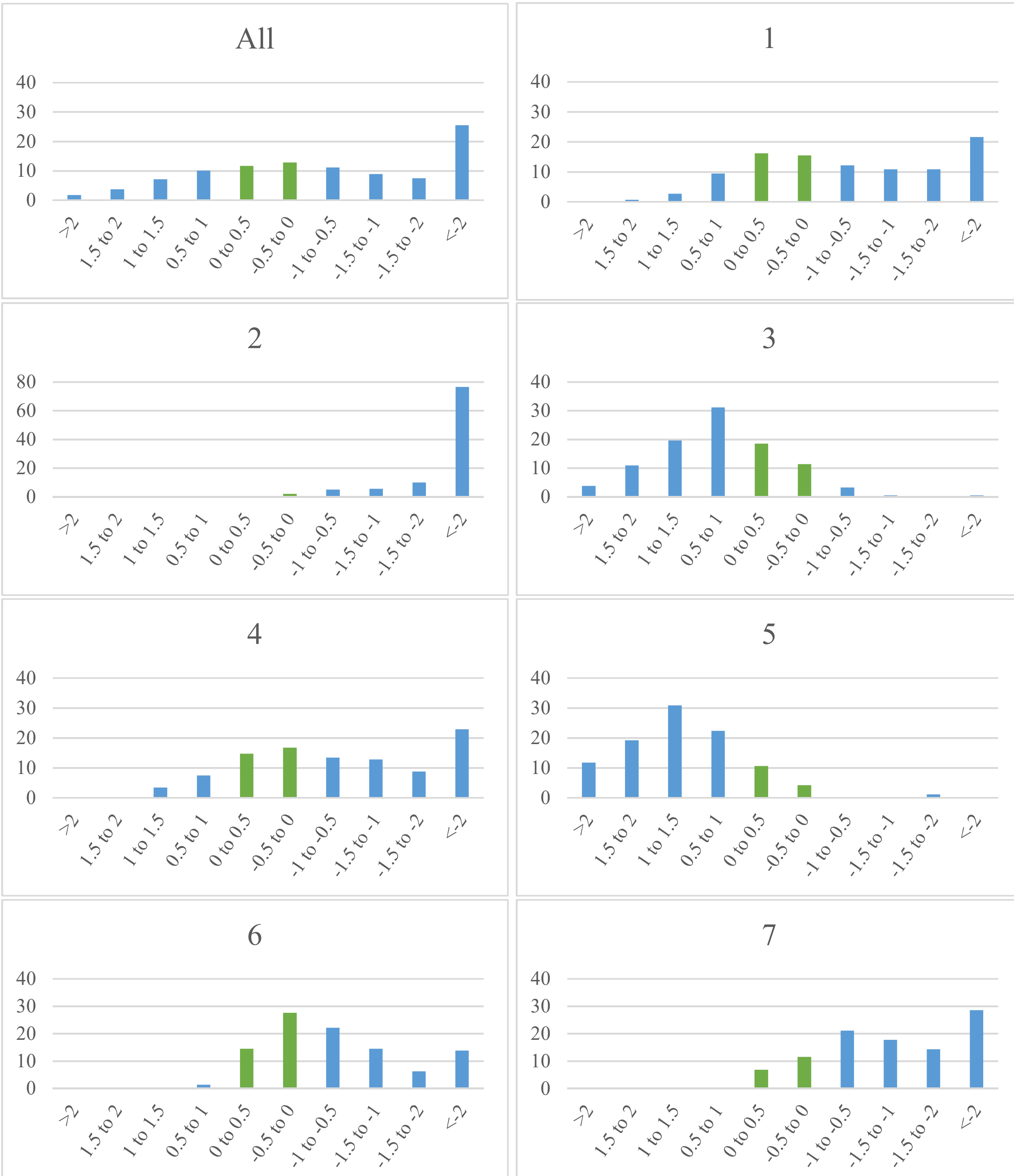

Figure 4.6. Residual for crude fat percentage of image intramuscular fat of overall result and each individual meat plant. Residual= crude fat percentage – image intramuscular fat. Green bars represent correct prediction (-0.5 < residual < +0.5)

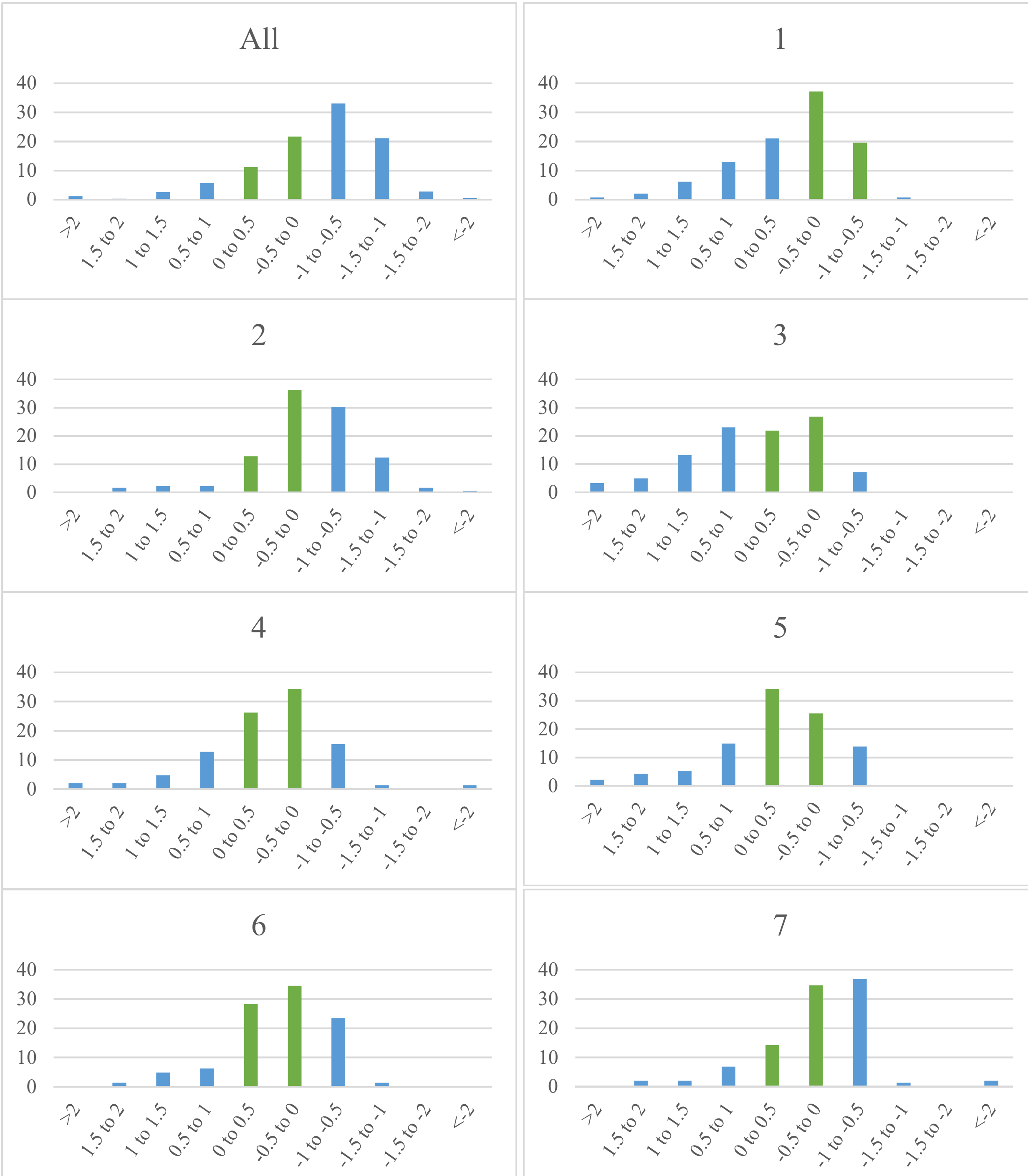

Figure 4.7. Residual for crude fat percentage of regression intramuscular fat of overall result and each individual meat plant. Residual= crude fat percentage – regression image intramuscular fat. Green bars represent correct prediction (-0.5 < residual < +0.5)

fat on surface as CF%, while our CVS is capable of automatic selecting the cleanest area as ROI, however, in each meat plant, there are different standard in trimming, one meat plant would demand a very cleaned, well-trimmed whole loin, while others may prefer minimal trimming, and this could increase the difficulty of ROI selection as ROI is a fixed area and could have chances of having unwanted subcutaneous fat or connective tissue counted as marbling; while trained grader could easily distinguish the difference, CVS would have to rely on the consistency of the cleanness of the surface of loin. 2. ROI position and size. It was noticeable that the positioning of ROI was not consistent as it relies on the consistency of cleanness of the loin lean surface, depending on the cleanness of the lean surface area, the ROI could be closer to the anterior rib or the posterior, which could affect the precision of our prediction. Also, due to the inconsistency of trimming or cleanness of the lean surface, it would restrict the size of ROI as the system would try to avoid subcutaneous fat or connective tissue automatically. This could further influence the precision of our prediction as the distribution of marbling through the whole loin could vary between each loin. 3. Using CF% of 2 chops to represent whole loin CF%, the distribution of marbling within the whole loin varies. between location. However, when switching to RIMF, the residual distribution shifts towards being more accurate. However, there is a slight overestimation (left skewness) of CF%. This could possibly be improved by ensuring that the regression model development data set has equal representation of samples in each CF% category. 4. Using equal amount of sample for each category. In our results, the residual distribution has shown to be left skewed, another possible reason could be due to the imbalance size of our sample, while category one represents over 80% of the sample and category five only 1%.

Conclusion

Neither of the prediction accuracies were satisfying, which suggests more research should be conducted to improve accuracies or to find the most ideal ROI on lean surface loin to represent the whole loin. When comparing results in prediction of CFC, CVS had an overall higher accuracy (58.50%) when compared to SMS (53.30%). The residual distribution of the RIMF was left skewed suggesting that CVS system tends to over-estimate CF% when comparing to SMS, however, the residual distribution was also more concentrated when comparing to SMS (95.22% vs 89.56%, residual within -1.5 and +1.5). Overall, this demonstrate that CVS has a better accuracy and precision when compared to subjectively grading marbling and the potential of application of using CVS as an objective measurement of CF% in both the industry and laboratory.

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CHAPTER 5. SUMMARY AND CONCLUSIONS

The studies conducted and presented in this dissertation have allowed for the opportunity to demonstrate the possibility of using computer vision system (CVS) as an objective measurement tool for predicting crude fat percentage (CF%) of pork. Through our series of experiments, we have moved from a system that was laboratory based to a system that is industry friendly.

In Experiment 1, we investigated the possibility of using CVS to predict CF% under a laboratory based environment. Samples were already trimmed of subcutaneous fat and connective tissue before image acquisition, to minimize the factors which could affect our imaging process and results. Our CVS though build precise, however, wasn't suitable for industrial purposes. When setting up a model for predicting CF%, 18 color features were used; stepwise regression and support vector machine were adopted. In result, while the support vector machine has shown to have higher accuracy when looking at categories (75 vs. 64 %) but stepwise regression had a narrower residual distribution (65 vs. 44 % between -0.5 and +0.5).

In Experiment 2, we upgraded our CVS to meet industry requirements. Instead of using a Charge Couple Device, we upgraded our camera to an industrial smart camera. Our lighting system was upgraded by introducing a 44-inch dome light system which was synded with the camera and image collection software instead of using two LED bar light build within a box. To improve portability, our system was built on a table with wheels for transport, which also allowed for constant distance between the camera and the sample. Instead of using Matlab software, Nivision was adopted, which is more common and compatible to the industry needs. When comparing accuracies to subjective marbling score (SMS), CVS has shown the potential of competing with SMS. A lower accuracy in anterior chop than posterior chop was noticed in both

SMS and CVS, which could be due to the variation of pork color, texture, and the transparency of marbling. Future research based on increasing model accuracy such as adding color features or texture features is warranted.

In Experiment 3, the same CVS system as Experiment 2 was used. However, the images were acquired within the meat plant and were of the whole pork loin instead of individual chops. Both the accuracy of CVS and SMS dropped compared to Experiment 2. When observing the surface of loin, it was noticed that there was a sizeable variation in color and marbling between the anterior and posterior ends of the loin. This, in conjunction with the online speed and the different standards of trimming between plants, could all potentially be factors that influenced both CVS and SMS. Similar results were also reported by Carpenter et al. (1961) and Faucitano et al. (2004), who observed extreme variation in IMF content and marbling at different loci of thoracic ribs from pig carcasses. In 2013, Hang et al. reported that the nonuniformity of sampling site has been observed in various studies and believes that could cause discrepancy in results in pork quality studies; it is also reported that using last rib had the great potential for prediction of CF% of the whole loin. Future research could focus on finding the most ideal anatomical sites that best correlate with the whole loin quality. Within the series of our study, the influence of CF% and pork lean color has been established, this suggest that further research such as adding color features or textures features to increase model accuracy and robustness is needed.

As a result, the possibility of using CVS as an objective measurement for CF% in pork has been established. While in the past pork has always been more driven by quantity over quality, more and more research has proven the importance of pork quality, and its impact on consumers' willingness to purchase and on pork export. Therefore, further research is warranted and this will forever be an area to explore as the demand of pork quality continues to increase.