

STUDY OF THE WOODY BREAST AND WHITE STRIPING CONDITIONS
AFFECTING THE PECTORALIS MAJOR MUSCLE OF BROILER CHICKENS:
PROCESSING PERSPECTIVE

A Dissertation

by

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Submitted to the Office of Graduate and Professional Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

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December 2019

Major Subject: Poultry Science

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ABSTRACT

Woody breast (WB) and white striping (WS) are meat quality issues affecting the *pectoralis major* muscle of broiler chickens. Three studies were performed to address WB and WS. The first study evaluated the effect of citrus fiber (CF), rice starch (RS), and sodium tripolyphosphate (STPP) in different combinations on the quality traits of WB. Treatments; Control) NaCl (0.83%) + STPP (0.45%), T1) NaCl (0.83%) + STPP (0.45%) + RS (1.5%), T2) CF (1.3%), and T3) CF (1.3%) + STPP (0.45%) were evaluated. Results indicated that RS+STPP, CF and CF+STPP could be used to improve marinade retention and cook loss yield in breast meat affected with the WB condition. Citrus fiber could be a potential clean label ingredient used as substitute of STPP in marinade brines. The second study investigated the effects on quality traits, acceptability, and instrumental texture of fresh chicken sausage and deli-loaves formulated with severe WB and/or normal (NOR) meat as follow: T1)100% NOR + 0% WB, T2) 75% NOR + 25% WB, T3) 50% NOR + 50% WB, T4) 25% NOR + 75% WB, and T5) 100% NOR + 100% WB. Data indicated that chicken sausage can be formulated with up to 75% WB without affecting the consumer perception towards the texture and cook yield of the product. The combination of 25%;75% normal and severe WB fillets could be a suitable option in the formulation of chicken deli-loaves without affecting the texture and yield of the final product. The objectives of the third study were to 1) to investigate the relationship between WS and WB, and 2) to propose a predictive model that uses growth production factors to investigate the incidence and severity of WS and

WB. Results indicated that there is a positive but moderate association between WS and WB and are jointly predictive of the severity of WS and WB. Variables such as age, live weight and sex were not as important as breast weight and species in the severity prediction of both myopathies. Potentially other factors not included in this study, may play a major role in the relationship of these myopathies.

DEDICATION

To my family

ACKNOWLEDGEMENTS

I would like to thank my committee chair, Dr. Coufal and Dr. Alvarado, and my committee members, Dr. Lee, Dr. Kerth, and Dr. Smith for their guidance and support throughout the course of this research.

Thanks also go to my friends and colleagues and the department faculty and staff for making my time at Texas A&M University a great experience.

Finally, thanks to my mother and father for their encouragement and to my fiancé Fernando for his patience and love.

CONTRIBUTORS AND FUNDING SOURCES

Contributors

This work was supported by a dissertation committee consisting of Dr. Coufal [advisor], Dr. Alvarado [co-advisor] and Dr. Lee of the Department of Poultry Science and Dr. Kerth and Dr. Smith of the Department of Animal Science.

The data analyzed for Chapter 4 was conducted in part by Ryan Travis.

All other work conducted for the dissertation was completed by the student independently.

Funding Sources

There are no outside funding contributions to acknowledge related to the research and compilation of this document.

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1. INTRODUCTION AND LITERATURE REVIEW

1.1. Introduction

Poultry meat is a popular protein around the world. Factors such as price, versatility, absence of cultural or religious obstacles, and nutritional properties make chicken highly consumed compared to other protein sources (Haley, 2001; Neth and Parker, 2018). In 2018, the per capita consumption of poultry meat in the US was approximately 41.78 kg. Consumption is approximately double when compared to beef and pork and is estimated to grow even more in the next few years (National Chicken Council, 2018). Therefore, to meet consumer demand for chicken, producers have improved production performance of live birds by improving nutrition, genetics, feed efficiency, farm practices and animal welfare (Petracci et al., 2014). According to the National Chicken Council (2017), the average number of days to raise a chicken was reduced from 63 in 1960 (1.52 kg) to 47 in 2016 (2.79 kg). As a result, chickens are marketed in less time and at about twice the weight compared to 57 years ago. Furthermore, chicken production has grown almost 900 percent since 1960, reaching a record 21 billion kg of ready-to-cook weight in 2015 (National Agricultural Statistics Service, 2016). This increase in consumer demand for chicken meat and the preference for breast meat in western countries, has also led to progressive changes in selection criteria toward broilers with high breast meat development. However, the selection for fast growing birds with high breast yield has resulted in an increase in breast muscle abnormalities such as deep pectoral muscle myopathy (green muscle), pale soft

exudative (PSE), and white striping (WS) and woody breast (WB) meat (Dransfield and Sosnicki, 1999; Petracci and Cavani, 2012; Kuttappan et al., 2012a; Petracci et al., 2013a; Sihvo et al., 2014).

Deep pectoral myopathy and PSE have been extensively investigated in broiler chickens in the last 20 years. Deep Pectoral myopathy (green muscle disease) develops in the pectoralis minor and is characterized by ischemic necrosis. This muscle is surrounded by an inelastic membrane and the sternum, and as a result, the muscle mass is not allowed to swell in response to the physiological changes occurring when muscles are exercised (Jordan and Pattison, 1998). In fast-growing strains, the increased pressure within the muscle restricts the blood supply and causes a necrosis of the muscle. This condition can affect just one or both pectoralis minor muscles (Petracchi and Cavani, 2012). On the other hand, PSE-like meat is characterized by pale color, soft texture and low water-holding capacity (WHC) mainly in the pectoralis major muscle (Barbut, 1997; Petracchi and Cavani, 2012). PSE-like meat is the result of the combination of fast postmortem pH decline (acid conditions) along with high carcass temperatures during slaughter. This combination leads to protein denaturation, which is associated with poor quality properties in the meat (Santos et al., 1994). Factors such as environmental temperatures (Cassens et al., 1975), pre-slaughter conditions (D'Souza et al., 1998), stunning methods (Backstrom and Kauffman, 1995), and chilling regimens (Offer, 1991) are associated with severity of PSE meat. According to recent publications, the selection for fast-growing strains has induced histological and biochemical changes and an

increased susceptibility to stress which can also be related with the PSE-like condition (Barbut et al., 2008; Sandercock et al., 2006).

In the past few years, two new abnormalities were discovered in the Pectoralis major muscle characterized by degenerative myofibrillar proteins called white striping and woody breast (Sihvo et al., 2014). White striping is characterized by the white striations with varying degrees of severity parallel to the muscle fibers (Kuttappan et al., 2009; Petracci and Cavani, 2012). Moreover, WS is not only a visual appearance issue, but also involves histological changes associated with muscle fiber degeneration and myopathic changes beneath the striation area resulting in reduced functional properties and changes in the fatty acid composition (Petracci et al., 2013a; Kuttappan et al., 2012b; Kuttappan et al., 2013a). Recently, it has also been observed that WS breast fillets can be accompanied by another type of muscle abnormality known as woody breast. Woody breast is characterized by the abnormal hardness throughout the fillet with different degrees of severity (Sihvo et al., 2014). The dramatic increase of WB meat during the last five years has become a challenge for the global poultry market. Kuttappan et al. (2017) reported that the incidence of WB increase as the birds get older and heavier. Recent research has shown that WB fillets have a different texture profile, less water holding capacity associated with lower higher drip loss, and lower marinade retention and cook loss when compared to normal fillets (Aguirre et al. 2018; Chatterjee et al., 2016; Soglia et al., 2017). In order to alleviate the current industry situation, it is necessary to further study alternatives to improve the quality properties of WB meat with respect to marination properties and further processed products. In addition, previous

research reports that WB and WS exhibit similar histological change; in this respect, it is imperative to understand the relationship between WB and WS. Recently, the Food Safety and Inspection Service (2017) sent disposition instructions for WB and WS poultry conditions. The FSIS Notice explains that the severe inflammatory tissues associated with WB and WS must be removed during inspection. These new dispositions plus the consumer's unwillingness to eat WB are causing an economic impact for the poultry industry (Kuttappan et al., 2012a).

Therefore, this study will investigate the WB condition in three different experiments: 1) Effect of different marinade ingredients on the quality traits and texture profile analysis of WB meat; 2) Quality traits and texture profile analysis of fresh sausage and deli-loaves chicken products formulated with different percentages of WB meat; and 3) Analysis of the relationship between WB and WS condition in chicken.

1.2. Mechanisms of muscle development

Muscle growth, composition, and metabolism are linked to meat quality through effects on yield, tenderness, and color (Brandeboung, 2013). There are two periods of muscle growth: prenatal or hyperplasia where the number of muscle fiber increases until set at hatch and postnatal or hypertrophy which happens post-hatch and is where preexisting muscle fibers increase in size. In the absence of injury, fiber number is maintained during this period as very little new muscle fiber growth occurs after birth. The adult myoblast cell population is made up of satellite cells, which are mesodermally derived stem cells responsible for all post-hatch muscle growth through hypertrophy

(Smith, 1963). Satellite cells are located between the basal lamina and the sarcolemma of mature muscle fibers. In normal chicken muscle, satellite cells are responsible for the maintenance of the fiber muscle by facilitating the regeneration of damaged muscle fibers. When satellite cells are activated in response to damage, they enter the cell cycle, increase their cell number through proliferation, and differentiate into multinucleated myotubes, which fuse with existing muscle fibers or create new fibers (Mauro, 1961; Moss and LeBlond, 1971; Montarras et al., 2005). Modern broiler lines have been selected for muscle growth through hypertrophy, thus having a larger fiber diameter compared to the standard strains. In addition, hypertrophic birds have a cross-sectional area three to five times wider than non-selected birds (Dransfield and Sosnicki, 1999).

1.2.1. Muscle tissue

There are three major types of muscle tissue in livestock, smooth muscle, cardiac muscle and skeletal muscle. Smooth muscle is mainly found in the wall of blood vessels, gastrointestinal track, uterine walls, and walls of the respiratory passage. Smooth muscle contraction is involuntary since it is controlled by the autonomic nervous system. Cardiac muscle is unique to the heart and has involuntary contraction. Skeletal muscle represents 35 to 65 % of carcass weight of meat animals and its contraction is voluntary since it is controlled by the nerves emanating from the spinal cord. In particular, skeletal muscle constitutes 40 to 50 % of the average body mass of an adult bird (Barbut, 2015). Skeletal muscle has a unique and highly organized structure which facilitates muscle movement. In addition, skeletal muscle is striated as a result of the abundant expression of contractile apparatus, proteins, and overall fiber composition.

These muscles are attached directly or indirectly to bones via ligaments, fascia (connective tissue), cartilage or skin (Aberle et al., 2001; Brandebourg, 2013).

1.2.2. Muscle structure

Within the skeletal muscle, each muscle is covered with a connective tissue sheath. The epimysium is around the whole muscle and is contiguous with the perimysium and the endomysium. The muscle is composed of muscle bundles of fibers that are surrounded by the perimysium and individual muscle fibers and surrounded by endomysium. The basic structural unit of skeletal muscle is the muscle fiber or also known as the myocyte, myofiber, or muscle cell. Muscle fibers constitute 75 to 92 % of total muscle volume (Aberle et al., 2001). Muscle fibers either from mammalian or avian skeletal muscle are single, long, multinucleated, unbranched, threadlike cells that taper slightly at both ends. Individual muscles are composed of variable number of fibers that run parallel to one another. Even though fibers can reach a length of many centimeters, usually they do not extend this entire length. These fibers, have different diameters 10 to 100 μm within the same muscle. Surrounding the muscle fibers there is a semi-permeable plasma membrane called the sarcolemma which is composed of proteins and lipids. The sarcolemma is relatively elastic to endure distortion that occurs during contraction, relaxation and stretching of the muscle. The sarcoplasm is the cytoplasm of the muscle fibers that contains lipid droplets, glycogen granules (energy reserve of muscle cells), ribosomes, proteins (e.g. myoglobin), non-protein nitrogen compounds, and a number of inorganic constituents (Aberle et al., 2001; Listrat et al., 2016).

Within each muscle fiber are myofibrils lined up in bundles, which occupy nearly the entire intracellular muscle fiber volume regardless of the species. Myofibrils constitute the contractile apparatus of the muscle fiber because it contains contractile units known as sarcomeres. The sarcomere is the repeating structural unit and it is also the basic unit where muscle contraction and relaxation occur. The distance from one Z-line to another comprises an individual sarcomere that consist of alternating thick and thin protein filaments giving skeletal muscle its striated appearance. (Alberts et al., 2008; Velleman, 2018). The thick filaments make up the A band of the sarcomere and predominantly consist of myosin. The thin filaments constitute the I band of the sarcomere and consist of actin, tropomyosin, troponins, tropomodulin, and nebulin, and is bisected by a Z line (Forrest et al.,1975). Tropomodulin caps the end of the thin filament near the M line and also determines actin filament length (Fowler et al., 1993). The H zone, pseudo H zone, and M line are other features in the myofibril that change in appearance according to the contraction state (Aberle et al., 2001).

Myofibrils contain more than 20 different proteins. From that list, myosin, actin, titin, tropomyosin, troponin, and nebulin represent more than 90 % of the overall myofibrillar proteins. According to their function, these proteins are classified as either contractile, regulatory or cytoskeletal proteins. The major contractile proteins are actin and myosin. Actin represents 20 % of the myofibrillar proteins. It is referred as G-actin because of its globular shape and has a diameter of ~ 5.5 nm. G-actin are linked together in strands to form a single fibrous molecule called F-actin. Two strands of F-actin are spirally coiled around one another to form a super helix which is characteristic of the

actin filament (Aberle et al., 2001). Myosin of the other hand, is a fibrous protein which represents 45 % of myofibrillar protein. Myosin contains two heavy chains (myosin heads) and two light chains (myosin tails). Tropomyosin and troponin constitute the major regulatory proteins. The role of regulatory proteins is to regulate actin and myosin interactions during muscle contraction. Tropomyosin makes up 5 % of myofibrillar proteins and is a rod-like protein that surrounds the helical structure of the actin filament. Troponin, likewise constitute 5 % of myofibrillar protein and lies within the actin filament along the tropomyosin strands. There are three types of troponin molecules: 1) Troponin C, that binds Ca^{++} , 2) Troponin I, inhibits adenosine triphosphate (ATP) and, 3) Troponin T, binds tropomyosin (Barbut, 2015). As for cytoskeletal proteins, titin (10 % of myofibrillar protein) and nebulin (4 % of myofibrillar protein) serve as a template or provide the scaffold for the alignment of myofilaments during the formation of myofibrils and sarcomeres (Judge et al., 1989; Aberle et al., 2001).

1.2.3. Muscle contraction and relaxation

Muscle contraction is the result of a chain of events. Muscle will contract or relax when they receive signals from the nervous system. Muscle contraction is the result of thousands to millions of sarcomeres moving in unison to produce tension (Barbut, 2015). The most comprehensive theory about muscle contraction is the sliding filament theory, which explains that in order for contraction to occur, the thick myosin filaments slide between the thin actin filaments towards the Z-lines (Huxley, 1958; Warriss, 2010). Because the actin filaments are anchored to the Z-lines, the sarcomeres shorten from both sides when actin filaments slide along the myosin filaments. Although it is known

as the “sliding theory”, myosin pulls the actin along its length. According to this theory, the sarcomeres shorten without the thick or the thin filaments changing in length. A contraction begins when the myosin head is energized via myosin-ATPase activity, where a bound ATP is hydrolyzed to adenosine diphosphate (ADP) and inorganic phosphate. This causes the myosin head to extend and can attach to a binding site on actin forming a cross-bridge. An action, called the “power stroke”, is triggered allowing myosin to pull the actin filament toward the M-line. Thereby shortening the sarcomere. The ADP and inorganic phosphate are released during the “power stroke”. The myosin remains attached to actin until a new molecule of ATP binds freeing the myosin to either go through another cycle of binding and more contraction or remain unattached to allow the muscle to relax. Muscle contractions are controlled by the actions of Ca^{++} . The thin actin filaments are associated with regulatory proteins troponin and tropomyosin. When muscle is relaxed tropomyosin blocks the cross-bridge binding sites on actin. When Ca^{++} ions levels are high enough and ATP is present, Ca^{++} ions bind to the troponin-C which displaces tropomyosin, exposing the myosin binding sites on actin. This allows myosin to attach to a binding site on actin forming a cross-bridge. Ca^{++} ions are stored in the sarcoplasmic reticulum and are released in response to signals from the nervous system to contract. Neurotransmitter molecules are released from a neuron and bind to receptors which depolarize the membrane of the muscle fiber. The depolarization of the sarcolemma occurs for an influx of Na^+ and efflux of K^+ for polarization reversal of the membrane. The electrical impulse travels down the T-tubules and opens Ca^{++} stores. Ca^{++} flow to the myofibrils where they trigger muscle contraction. The repeated

formation and breaking of cross bridges results in sliding of the thick filaments towards the Z-lines and, hence, sarcomere shortening (Aberle et al., 2001; Alvarado and Owens, 2006; Sayas-Barbera, 2010). When muscle fibers contract in unison, a muscle can produce enough force to move the body. During the relaxation phase, signals from the nerve diminish, the sarcolemma and T-tubules are re-polarized preparing for the next signal, Ca^{++} has to be resequenced into the sarcoplasmic reticulum, cross bridges are broken and cannot reform and ATP must be regenerated. The tropomyosin molecules cover actin binding sites. Finally, sarcomeres return to their resting state (Barbut, 2015). Repolarization of the sarcolemma is established by the Na^{+}/K^{+} pump transporting Na^{+} out of the cell and K^{+} into the cell and resequencing of Ca^{++} is pumped back into the sarcoplasmic reticulum via ATPase (Alvarado and Owens, 2006).

1.2.4. Conversion of muscle to meat

In a living animal, organs, temperature, pH, oxygen, and CO_2 concentration are in a dynamic state of equilibrium known as homeostasis. During the bleeding process, at harvest, approximately 50 % of the blood is removed from the carcass, while the rest of the blood is contained within the vital organs and muscles. Blood removal stops communication between muscles and vital organs (Braden, 2013; Barbut, 2015). Since the bloodstream transports nutrients and oxygen, the oxygen is depleted and energy metabolism changes from aerobic (TCA cycle) to an anaerobic pathway to provide energy to the muscle for a period of time. In a living animal, the lactic acid produced by anaerobic metabolism is transported from the muscle to the liver, where it is re-synthesized into glucose and glycogen, or to the heart, where it is metabolized to CO_2 and

water. Since the bloodstream is not able to remove waste products, lactic acid remains in the muscle and its concentration increases during the time postmortem. Lactic acid buildup continues until the glycogen stored in the muscle is depleted. Because of the buildup in lactic acid, the pH of the meat gradually starts to decrease from 7.4 (living muscle) to ~5.8 (Aberle et al. 2001; Braden, 2013). In poultry breast muscle, the drop in pH occurs more than twice as fast as it does in beef and pork. pH decline is one of the most significant postmortem changes that can influence meat quality. Another change postmortem is rigor mortis which is the Latin for stiffness of death. During the earliest postmortem phases, creatine phosphate is used to form ATP. Since creatine phosphate is depleted faster, ATP starts to decrease. ATP is needed for the muscle to relax, since there is no ATP, the muscle will become inextensible as a result of the formation of actomyosin bonds. Once the muscle becomes inextensible, rigor mortis is complete (Aberle et al., 2001). After certain postmortem storage time, muscle becomes flexible again in a process called “resolution of rigor mortis”. However, this reduction in muscle tension is not associated with the actomyosin bonds being broken down. Rather the decrease in tension is the result of proteolytic degradation of sarcomere component, Z-lines, myofibrillar proteins, connective tissue and loss of structural integrity (Aberle et al., 2001; Warriss, 2010; Barbut, 2015).

1.2.5. Muscle proteins

Skeletal muscles are composed of 75% water, 20% protein, 1–10% fat, and 1% glycogen. There are more than 50 different types of proteins but only five represent the major proportion. According to their solubility, proteins are categorized into three major

groups; sarcoplasmic, myofibrillar and stromal proteins (Asghar et al., 1985). Sarcoplasmic proteins (soluble) represent approximately 30% of the muscle proteins. They are located in the sarcoplasm and consist of myoglobin, hemoglobin, cytochromes, glycolytic enzymes and creatine kinase. Myofibrillar proteins (insoluble) represent 55% of the muscle proteins. These proteins are considered as the building blocks of muscle and also contain the contractile proteins actin and myosin as well as tropomyosin, troponin, C-protein, α -actinin and β -actinin. The stromal proteins represent approximately 12% of the muscle. They consist of collagen, elastin which are part of the connective tissue, and mitochondrial proteins. They are not water-soluble or salt-soluble proteins. These proteins form structural components including membranes (surround cells), muscle bundles, ligaments, tendons and joints (Barbut, 2015).

1.3. Meat quality

1.3.1. Meat color and pH

Color is the first quality trait considered by consumers when making fresh meat purchasing decisions because they use it as an indicator of product spoilage and wholesomeness (Mancini and Hunt, 2005). Meat color is influenced by the amount of myoglobin and the oxidative status of the heme iron found in them. Although, there are several intrinsic and extrinsic factors that can affect meat color, including but not limited to, species, nutrition, age, sex, physical activity, type of muscle, antemortem and post mortem changes, processing methods, use of non-meat ingredients (e.g. nitrite), lightning conditions, and packaging materials. In particular, meat color is affected by

myoglobin content, muscle fiber orientation, space between the fibers and pH. Chicken breast meat is predominantly composed of white fibers, therefore having low myoglobin content and light gray color. Whereas, thigh meat is mainly composed of red fibers and appears dark (Barbut, 2015).

Myoglobin is a sarcoplasmic protein that determines meat color via its centrally located heme iron. Myoglobin is composed of two major parts: the protein component consists of a globular protein and the non-protein component consist of the heme ring, which has an iron molecule in it's center which is responsible for binding molecules such as oxygen and water. The oxidation state of the iron molecule and the compounds attached to the ion determine the shade of red color (Aberle et al., 2011; Barbut, 201). The purple-red color characteristic of a freshly cut surface is known as myoglobin and resembles the absence of oxygen together with ferrous heme iron (Fe^{++}). Bright pink color or oxymyoglobin is the result of oxygenated myoglobin and the iron molecule in its reduced ferrous state (Fe^{++}). Brown color or metmyoglobin occurs, as a result of the oxidation (electron loss) of the heme iron from ferrous (Fe^{++}) to a ferric (Fe^{+++}) often associated to the release of oxygen radical (Sato and Shikama, 1981; Wallace et al., 1982). Once the meat is exposed to heat as a result of cooking, the meat pigment color is usually grey/brown. Heat denatures the globular protein portion of myoglobin and the heme ring is usually separated from the myoglobin and adds to the non-heme pool in meat. The fully denatured myoglobin becomes the cooked pigment or know as denatured metmyoglobin. As a result of denaturation, meat changes to a more opaque structure and reflects more light. For example, in the case of cooked chicken breast (lower myoglobin

content) the L* (lightness) values usually increase from an average of 52 to 82 which represents approximately 60 %. In addition, breast color becomes more yellow going from an average of 6 to 14 (Barbut, 2015).

The color attributes tend to be strongly associated with the pigment myoglobin, whereas the lightness is related to structural attributes of the muscle and together these determine the reflected light which is visualized. In addition, when light comes in contact with the surface of the meat, it can either be reflected, absorbed or scattered (Hughes et al., 2014). For example, PSE-affected breast meat usually has a great amount of extracellular water resulting in many reflecting surface areas therefore, it will have limited absorption capabilities. On the other hand, DFD (dark, firm, dry) meat has a great amount of intracellular water therefore, white light reflection is minimized, and color absorption is enhanced.

Muscle pH decline and ultimate muscle pH have considerable effects on meat color. If the animal has antemortem stress, glycogen storage can be depleted prior to slaughter, which results in low lactic acid production and therefore pH drop will be minimal causing a high ultimate pH of 6.5 and 6.8. This condition is known as dark, firm, and dry (DFD). On the other hand, the fast drop in pH (5.4-5.5) while the meat temperature is still high (e.g. $>35^{\circ}\text{C}$), can cause protein denaturation resulting in low water holding capacity, this condition is known as pale, soft, and exudative (PSE) meat.

1.3.1.1. Instrumental surface color evaluation

Two methods of measuring color have been used by researchers: consumer and trained panels for visual methodology, and surface reflectance for instrumental techniques (Mancini, 2013). Regardless of the methodology, several considerations should be considered in order to have meaningful results. In particular, researchers must ensure that a representative number of samples are obtained, samples should be prepared and analyzed in the same manner, the location within a muscle should be considered, and lightning type must be maintained from sample to sample, panelist to panelist and replication to replication (O'Neill et al., 2003; Mancini, 2013). Even though visual color evaluation is closer to what consumers see, researchers prefer the instrumental color evaluation because of its rapid and non-destructive nature. The most common instrument to measure surface color is the CIE (Commission Internationale de L'eclairage) L^* , a , b^* , which also provides more reproducible data compared to other methods (Hulsegee et al., 2011; Garcia-Esteben et al., 2003). This commercial spectrophotometer is equipped with a stable light source (i.e. xenon) to illuminate the surface after the instrument has been calibrated with a white plate (Barbut, 2015). Following is the definition of the variables:

L^* = indicate lightness, 100= white, 0= black, increased values are lighter

a^* = measure redness, positive values are red, increased values are more red

b^* = measure yellowness, increased values are more yellow

Another important consideration when utilizing reflectance methodology to characterize surface color, is to cover the tiles or glass plates with the same packaging film as the meat samples (Mancini, 2013).

1.3.2. Water holding capacity (WHC)

Muscle contains 75% water (Aberle et al., 2001). Water holding capacity has been defined as the ability of the meat to retain its water during application of external forces such as cutting, heating, grinding or pressing is an important characteristic of fresh meat as it affects both the yield and the quality of the final product (Huff-Lonergan, 2006). This characteristic is often measured as drip, purge, and cook loss (Honikel and Hamm, 1994). It has been estimated that product weight loss due to purge can average as much as 1 to 3% in fresh retail cuts and as much as 10% or more in poor quality (PSE) meat products (Offer and Night, 1988a; Melody et al., 2004). According to Savage et al. (1990), purge can contain approximately 112 mg of protein per millimeter of fluid; mostly water-soluble sarcoplasmic proteins. Myoglobin is a sarcoplasmic protein that provides the primary pigmentation for meat, therefore excessive water loss can lead to meat color fading which can cause consumer rejection and huge economic losses (Brewer et al., 1998). Thus, WHC is one of the most important quality attributes specially in fresh meat products.

Water is a dipolar molecule attracted to charged molecules similar to myofibrillar and stromal proteins, and polar molecules without charges. Water is distributed in three different muscle compartments known as bound, immobilized and free water (Aberle et

al., 2001). Bound water represents only 4 to 5 % of the total water in the muscle and is located strongly attached and bound within the myofibrillar proteins even during the application of severe mechanical or physical force (Fennema, 1985; Offer and Knight, 1988b). Immobilized water represents the largest portion of water approximately 80% of the volume of the muscle cell; however, the amount of this water depends on the amount of physical force exerted on the muscle. Although, this type of water is not very mobile among compartments, it can be removed by conventional heating and converted into ice during freezing (Fennema, 1985). Immobilized water is most affected by the rigor process and the conversion of muscle to meat (Offer and Knight, 1988b). Therefore, the goal of meat processors is to maintain as much of this immobilized water as possible in the meat (Keeton and Osburn, 2010). In addition, this type of water is held within the myofibril by either steric (space) effect and/or by attraction to the bound water, but is not bound to with myofibrillar proteins. The last muscle water muscle compartment is free water, which represents less than 10% of the total water found in pre-rigor meat, flows from muscle tissue unimpeded, and is held by weak surface forces (Huff-Lonergan, 2006).

1.3.2.1. Net charge

After rigor mortis takes place, the buildup in lactic acid causes the pH to decline. As the pH approaches the isoelectric point (~ 5.0 to 5.4) of major proteins such as myosin, there is a reduction of reactive groups available for water binding to the protein. This happens because the number of positive and negative charges are equal (net charge is zero), thus they tend to be attracted to each other causing a reduction of space within

the myofibril and only the left over are available to attract water (Offer, 1991; Hamm, 1986). On the other hand, when the pH is below the isoelectric point, there is an excess of positive charges, whereas when the pH is higher than the isoelectric point, there is an excess of negative chargers causing an increase attraction to water (Aberle et al., 2001).

1.3.2.2. Steric effect

Myofibrils contain an average of 85% of the total water in the muscle. It is estimated that one-third of water loss is due to pH and two-thirds due to steric effects. As muscles go into rigor cross-bridges form between the thick and thin filaments, thus reducing available space to reside (Offer and Trinick, 1983). This decline in filament spacing may force sarcoplasmic fluid from between the myofilaments to extra myofibrillar space. Additionally, during rigor development sarcomeres can shorten, further reducing the space available for water within the myofibril (Honikel et al., 1986). The stearic effect has been shown to have a direct proportion to the breakdown of ATP postmortem. The breakdown of ATP and the protein interactions associated with rigor mortis are largely responsible for the formation of a right network within the contractor proteins. Divalent cations such as Ca^{++} and Mg^{++} can combine with negatively charged groups of the protein, which tend to pull the protein chains closer together and prevents those reactive groups that are available from binding water (Aberle et al., 2001).

1.3.3. Meat tenderness

Tenderness is the most important contributor to eating satisfaction according to a consumer survey (Miller et al., 1995). Additional evidence suggested that consumers are

willing to pay premiums for guaranteed tender products (Shackelford et al., 2011). In particular broiler breast meat requires a postmortem aging time of 4 to 6 h before deboning to obtain more tender fillets (Lyon et al., 1985). Considering the time consuming and storage space of this practice, electrical stimulation (Dickens et al., 2002), and post-chill flattening (Lyon et al., 1992) have been used to reduce the need for postmortem aging time while providing tender chicken breast (Dickens et al., 2002). Moreover, there are several physical and chemical properties of muscle that determine meat tenderness. The primary components that contribute to meat tenderness are connective tissue amount and quality, sarcomere length, and myofibrillar protein degradation, however it is important to highlight that the myofibrillar component does not apply to poultry meats (Whipple et al., 1990; King et al., 2009).

1.3.3.1. Connective tissue

Connective tissue is composed of collagen, which provides structural support to muscles and transfer force generated during contraction to the skeleton. There are three basic fibrous layers of connective tissue around the muscle are epimysium, perimysium, and endomysium, which are known as connective tissue proper (Aberle et al., 2011). The association between collagen and tenderness depends on the type and amount present in the muscle. The type of collagen is important to determine the solubility of the tissue. The relative insolubility and high tensile strength of collagen fibers result from intermolecular cross-linkage. These cross-linkages are few in number and more easily broken in young animals. As the animal gets older, the number of cross-linkages increase and become stable, which reduces collagen solubility during heating (Avery et

al., 1996), and becomes less soluble as the animal ages which increases toughness (Shorthose and Harris, 1990). On the other hand, in young broilers the cross-links have not formed yet, therefore, the collagen present is soluble and will eventually melt during the cooking process (Owens and Meullenet, 2010). Furthermore, collagen content has not been an issue in young broilers due to early slaughter ages. However, the collagen content in modern young birds selected for high breast development is increasing, thus changing the texture profile (Velleman, 2015).

1.3.3.2. Sarcomere length

The contractile state of muscle is another important factor affecting meat tenderness. Shorter sarcomeres are associated with greater toughness caused by the increased overlap of myofibrillar filaments (Marsh and Leet, 1974), whereas muscles with longer sarcomeres result in more tender meat (Herring et al., 1967; Weaver et al., 2008). Shear values are often used to measure meat tenderness. Higher shear values are associated with tougher meat and lower shear values are linked to tender meat. In poultry meat higher shear values haven been associated with deboning time. Mehaffey et al. (2006) reported that regardless of age, deboning at 2 h postmortem resulted in higher shear values, pH values, and L* values compared to breast meat deboned at 4 h postmortem.

1.3.3.3. Myofibrillar protein degradation

After rigor mortis occurs, meat tenderization will increase over time. Meat tenderization is the result of endogenous proteolytic inhibitor calpastatin and μ , and m-

calpains (Kerth, 2013). In particular, μ -calpains have been found to produce myofibrillar degradation during postmortem storage (Koochmaraie, 1996). Several studies suggest that postmortem changes such as loss of Z-disk and sarcomere integrity are observed during refrigeration storage as a result of proteolytic degradation of myofibrillar proteins. The proteins observed to be degraded are mostly cytoskeletal including troponin-T, titin, nebulin, desmin, vinculin, filamin, synemin, and dystrophin (Taylor et al., 1995; Koochmaraie, 1996). However, it is notable that the major contractile proteins, actin and myosin, are unaffected during postmortem storage.

1.4. Meat Processing

A growing demand for convenient and ready-to-eat products has increased poultry processors' interest in developing consumer-oriented, value-added meat products. The final quality of processed products depends on both the selection of raw materials and the processing techniques. Processed meat products are defined as those products in which properties of fresh meat have been modified using one or more procedures including grinding, chopping, marinating, seasoning, altering of color or heat treatment (Aberle, 2001). Comminuted products consist of raw meat materials that have been reduced into small pieces, chunks, chips or flakes. In particular, sausage is classified as a comminuted seasoned meat product. Some advantages of this process are improved uniformity of product (uniform particle size), improved distribution of ingredients, and an increase in tenderness.

Temperature is an important factor in the quality of the final product, because excessive high temperatures can denature proteins leading to reduce stability of meat batters. One of the techniques implemented several hours before batter production, is preblending, this allows additional time for protein solubilization and swelling to occur. On the other hand, formed products such as deli-loaves, which are often sliced prior to merchandising, are placed in casings (i.e. fibrous casings) after comminution and blending, and then cooked to set their shape. Upon heating, the protein matrix gels and soluble proteins denature and coagulate to convert the product from semi-fluid to elastic-solid (Aberle, 2001; Petracci et al., 2013b).

1.4.1. Marination methods

Marination has been widely used by the meat industry to increase yield and improve flavor, tenderness, and product shelf life in fresh meat (Xargayo et al., 2001). There are three methods of marination: 1) Immersion, which is the oldest and is mostly used at home or in small operations, consists of submerging the meat in the marinade and allowing the ingredients to penetrate the meat through diffusion with the passage of time. However, it does not provide a uniform distribution of the ingredients and it's time consuming (Xargayo et al., 2001); 2) Vacuum-tumbling or massaging, which is widely used in all types of operations from a butcher shop to a food processing facility, subjects the product to agitation, which helps disrupt tissue structure and improve the distribution of the marinade ingredients. Bringing salt into contact with salt-soluble proteins (actin and myosin) results in a greater water binding and solubilization of proteins. Tumbled or massaged products also develop a protein exudate that binds meat chunks together when

later formed into deli-type products. Products also are more tender because of the hydrated state of proteins and the loss of structural integrity (Aberle, 2001; Smith et al., 2001); 3) Multineedle injection marination, is mostly used in in-line operations, the needles or probes are inserted and the marinade is injected as the probes are withdrawn, spreading the marinade throughout the piece (Smith, 2001).

1.4.2. Functional Ingredients

Functional ingredients improve texture and water holding capacity in raw and cooked meats. Functional ingredients also can reduce the effect of natural quality variability and at the same time can provide more flexibility for the introduction of new products to meet consumer demands and for the optimization of cost formulations (Petracci et al., 2013). Different types of functional ingredients in the form of inorganic salts such as sodium chloride and phosphates, and organic compounds mainly from plant and animal origins have been introduced to meet the consumer demands. Salts are added to enhance the functionality of myofibrillar proteins, whereas organic compounds are added to retain water/fat and texture modulation alongside myofibrillar proteins (Lamkey, 1998). Sodium chloride and phosphate, in particular sodium tripolyphosphate (STPP), are widely used by the poultry industry and the combination of both can improve protein functionality.

1.4.2.1. Sodium chloride

Sodium chloride (NaCl) is widely used in the food industry because of its multifunctional properties. This ingredient is used to enhance flavor, increase WHC and

to improve texture by solubilization/extraction of salt-soluble myofibrillar proteins. Moreover, NaCl also increases microbiological stability. Currently, there is no regulation for the use of NaCl, however a concentration of 0.65 to 1.6 percent is used in the majority of poultry meat formulations (Petracci et al., 2013c). According to Feiner (2006) and Hamm (1986), the theory about the role of NaCl in improving the WHC in meat products starts with the dissociation of NaCl into sodium (Na^+) and chloride (Cl^-), then Cl^- ions are absorbed more strongly than Na^+ ions to positively charged groups of myosin. Binding of chloride ions to myosin and actin filaments increases the electrostatic repulsive forces between fibers, causing unfolding of the proteins structure matrix and expanding the spaced between actin and myosin.

1.4.2.2. Phosphates

Phosphates are salts of phosphoric acid available in different chemical types. Tripolyphosphates (STPP) represent more than 50% of the phosphates used by the meat industry, and are used to improve WHC, tenderness, juiciness, prevent pigment oxidation and overall enhance flavor by changes in ionic strength, pH, and stability of actomyosin (Aberle, 2001; Petracci et al., 2013b). Currently, STPP are regulated and can be used up to 0.5% in the final product. The mode of action of phosphates starts with dissociation of the acto-myosin complex (formed during rigor mortis) by sequestrated Ca^{++} and magnesium Mg^{++} cations. Then, calcium makes bridges between actin and myosin during the contraction of the muscle where phosphates have the ability to breakdown these bridges (Feiner, 2006). Following this, phosphates cause a pH increase which increases the gap between ultimate pH and isoelectric point. Phosphates increase

the electrostatic repulsive forces which expand the spaces between actin and myosin allowing for more water to entrap in these gaps (Barbut, 2002). Moreover, phosphates increase ionic strength which lead to more muscle fiber swelling and protein activation (Feiner, 2006).

1.5. Recent muscle abnormalities affecting the *pectoralis major* muscle

Genetic selection in broilers has led to muscle fibers hypertrophy, which alters its structural, functional and metabolic characteristics. White striping (WS) and woody breast (WB) are recent meat quality issues around the world found in fast growing heavy-broilers. It is also not clear whether WB and WS share a common etiology, but they are often together in the same fillet (Kuttappan et al., 2012a; Lorenzi et al., 2014; Velleman, 2015). In the market, you can often find WB fillets with the presence of WS at the same time, and you can find normal fillets with the presence of WS. Several studies reported that the incidence of these conditions have a negative impact on meat quality, in fact, they could result in an estimated economic loss of more than \$200 million/yr (Mudalal et al., 2014; 2015; Bowker and Zhuang, 2016; Kuttappan et al., 2016).

1.5.1. White Striping

White striping is visually characterized for having white striations running parallel to the muscle fibers. The categorization and scoring system consist of: normal (score 0), without any distinct white lines; moderate (score 1) presenting white lines <1 mm thick; and severe (score 2) exhibiting white lines >1 mm thick (Kuttappan et al.,

2009). According to Trocino et al. (2015), fillets affected by moderate white striping condition are usually marketed as whole muscle to consumers, while severe cases are downgraded and used in the manufacturing of further processed products. In addition, WS with different degrees of severity is also present in chicken tenders, thighs and drumsticks (Bowker et al., 2018; Kuttappan et al., 2013).

1.5.1.1. Incidence and severity

The incidence of WS has been reported in countries including Italy, France, Spain, Brazil and the United States, where fast growing birds are used (Kuttappan et al., 2013; Petracci et al., 2013a; Lorenzi et al., 2014; Alnahhas et al., 2016). Lorenzi et al., 2014 studied the incidence of WS in medium (2.2 to 3.0 kg) and heavy (3.8 to 4.2 kg) broiler chickens under commercial conditions in Italy and reported that all commercial hybrids used in the study had some degree of WS, although there was more prevalence (60.3%) of WS breast on heavy birds. In addition, there was a significantly higher rate of moderately white-striped breasts in males than in females (31.3% vs. 21.7%). In the US, Kuttappan et al. (2013b) also reported that 55.8% of broilers between 59 to 63 d of age presented some degree of WS under experimental conditions. Petracci et al. (2013) found 12% of WS in lighter birds (~2.75 kg) slaughtered from 45 to 54 d of age. Other authors studied the incidence of WS in standard and high breast yield and found that 74.5% of the fillets used in the study presented WS at day 46 regardless of the genotype. It was hypothesized that the growth rate was so high in both genotypes to induce a similar white striping occurrence (Trocino et al., 2015). On the other hand, Kuttappan et al. (2013a) reported no significant effect of gender on WS occurrence, but they observed

that females had a higher rate of normal breasts whereas males showed a higher rate of severely WS breasts. These effects were attributed to differences in bird weight, suggesting that the increase in growth rate increased the incidence and severity of WS. Within this context, Reedy-Griffin et al. (2018) found that the onset of WS is at day 16 and the onset of WB is at day 21, moreover, at day 46 almost all of the broilers show degenerative fibers regardless of genotype and gender, therefore confirming the hypothesis that both conditions increase in severity as the birds get older and heavier.

1.5.1.2. Meat quality

White striping has been shown to have a negative impact on meat quality. Breast meat with the presence of either moderate or severe WS exhibit greater weights and cranial thickness compared to normal fillets (Bowker and Zhuang, 2016; Kuttappan et al., 2012b;2013b; Lorenzi et al., 2014). Color (L^* , a^* , and b^*) and pH are also different, the pH of the meat increased as the severity of WS increased and color (a^* and b^*) decreased (Trocino et al., 2015); however, results are not always consistent among studies (Petracci et al., 2013a; Mazzoni et al., 2015; Mudalal et al., 2015). Moreover, several studies with intact and sub-samples have suggested that the WS myopathy has a negative impact in WHC. Fillets with WS condition showed increased cook loss percentage in non-marinated and marinated meat, as well as, decreased in marinade uptake and retention and overall product yield (Petracci et al., 2013a; Mudalal et al., 2014; 2015). Contrary to those studies, Bowker and Zhuang (2018) reported that WS fillets did not have a major impact on WHC characteristics. It was hypothesized that discrepancies in the results between studies may be due to the facts that WS is not

evenly distributed throughout the fillet and the different sampling location. In this regard, the same authors developed another experiment to demonstrate that the effects of WS on WHC were influenced by the anatomical location of sampling within the fillets. The results of the study showed that samples taken from the ventral surface of the cranial ends of the fillets exhibiting moderate and severe WS had decreased marinade uptake, increased cook loss, and decreased final yield compared to normal fillets, whereas, samples taken from the dorsal surface of the cranial ends of the fillets showed no negative effects on WHC. Therefore, WHC measurements were most affected on the cranial-ventral surface of breast fillets containing WS.

1.5.1.3. Sensory Analysis

It is well known that visual appearance is one of the most important sensory attributes used by consumers to measure the quality of food products. In order to understand the consumers perception towards WS, Kuttappan et al. (2012a), developed a consumer sensory analysis with 75 subjects to evaluate the preference and willingness to buy breast fillets affected with different degrees of WS. Filet and tray pack pictures where shown to the consumers on a computer screen, results showed that there was a decrease in consumer acceptance for breast fillets when the severity of WS increased. Moreover, 50% of the consumers disliked the visual appearance of severe WS, 22 % moderate, and 11% normal. Consumers were more willing to buy tray-packs containing normal meat than tray packs containing moderate or severe WS. These results suggest that the consumers were able to detect the occurrence of WS and perceived it as a negative attribute.

Sanchez Brambila et al. (2016) performed a sensory analysis of fillets affected by different degrees of WS and suggested that more effort is needed to compress, break and chew breast fillets with severe WS compared to normal and moderate fillets. The hypothesis of the different texture was attributed to the increased collagen (Petracci et al., 2014) and connective tissue (Kuttappan et al., 2013b) present in broiler breast meat. Furthermore, Tasoneiro et al. (2016) developed a descriptive conventional profiling sensory analysis of WS and WSWB, and revealed that off-odors in WS fillets had a higher overall intensity compared to normal ones, whereas WSWB fillets reached intermediate scores. Also, no differences in off-flavor, taste and texture were found between normal and WS fillets. In the same study, although only toughness was significantly different in WSWB fillets, all the other attributes in the sensory profile showed a detrimental effect on meat quality, nutritional quality and texture perception compared to normal and WS fillets. Thus, the combination of both myopathies in the same fillet may represent a major problem.

1.5.2. Woody Breast

The woody breast condition is characterized for being hard to the touch, pale in color, along with having a yellow viscous fluid on top of the breast fillets, and small hemorrhages (Dalle-Zotte et al., 2014; Sihvo et al., 2014). The WB categorization is done by tactile evaluation of hardness which consist of: normal (score 0), fillets are flexible throughout; mild (1), fillets are flexible mainly in the cranial region; moderate (score 2), fillets are hard throughout but flexible in the mid to caudal region; and severe

(score 3), fillets are very hard throughout from cranial region to the caudal tip (Tijate et al., 2016).

1.5.2.1. Incidence and severity

Reedy-Griffin et al. (2018) characterized the muscle development changes that occur with age/growth in commercial broilers and reported that the first documented case of WB was at day 23, and more severe characteristics were observed from day 30 onwards including thick white striations. Trocino et al. (2015) and Kuttappan et al. (2017) reported that the incidence of higher scores of WB increased with age and body weight. Moreover, fillet depth, bottom fillet height, and yield increased the probability of the muscle of having more severe WB (Mudalal et al., 2014; Kuttappan et al., 2017). Furthermore, the occurrence of WB was affected by gender, doubling from females to males (Trocino et al., 2015).

1.5.2.2. Meat quality

Meat quality of affected breast muscles have been of great concern for the poultry industry. Overall, WB and WS/WB fillets exhibits significantly higher pH compared to normal fillets regardless of the sampling method (Soglia et al., 2015; Kuttappan et al., 2017). Previous research has indicated that a pH close to the isoelectric point ~5.0 of muscle proteins leads to muscles with lower WHC, whereas, muscles with high final pH display better abilities to hold water (Hamm, 1961). However, the ultimate pH of fillets affected by either WB and WS or both conditions in the same fillets is higher compared to normal. In addition, WB WHC is significantly lower in non-

marinated and marinated fillets (Petracci et al., 2013a; Soglia et al., 2015; Kuttappan et al., 2017; Aguirre et al., 2018). Furthermore, the higher ultimate pH in WB fillets can be associated with a decrease in the glycolytic potential and altered energetic status and metabolic pathways found in previous studies (Zambonelli et al., 2016). In this regard, the severe degeneration of the muscle tissue in these myopathies would explain the poor WHC and the lack of correspondence between this parameter and pH (Dalgaard et al., 2018).

Among WS and WB, WB fillets were more affected and displayed particularly poor functionality which is expressed in higher drip loss; lower marinade uptake and retention; and overall lower cook yield (Mudalal et al., 2014; Tijare et al., 2016; Dalle-Zotte et al., 2017; Kuttappan et al., 2017; Aguirre et al., 2018). Soglia et al. (2015) evaluated the performance of WB and WS/WB (samples from the middle part) during vacuum tumble marination (15% wt/wt, 7.6% NaCl, 2.3% STPP) and water bath cooking, and found that marinade uptake was lower and cooking loss was higher compared to normal fillets. On the same line, Bowker et al. (2018), assessed the influence of WB on the marination (vacuum tumbled, 20% wt/wt, 5% NaCl, 3% STPP) and cooking (combi-steam oven) performance of the ventral (skin side) and dorsal (bone-side) breast fillets and reported even though there were negative effects of the myopathy on marination uptake/retention and cook yield, they seem to be less severe in the dorsal portion of the fillets. This is consistent with previous observations, where injected marinated (15% wt/wt, 0.55% NaCl, 0.48 STPP) individual fillets had lower marinade retention and higher cook loss regardless of the cooking method (flat-top grill

vs convection oven). Summarizing the results, it is evident that WB had a lower ability to hold water irrespective of the sampling methods.

Another important meat quality change in WB fillets is surface color (L^* , a^* , and b^*) and appearance. Zhuang and Bowker (2018) investigated the cooked color of normal and severe WB fillets as well as the effect of marination and frozen storage on the WB-induced discoloration and reported that WB cause a significant discoloration on the ventral surface of cooked breast meat. As a result, fillet surface was darker, redder, and yellower than the normal ones. In addition, discoloration of WB fillets was not eliminated by marination or freezing-thawing. According to Dalle Zotte et al. (2017) and Kuttappan et al. (2017) there is a positive relationship between the WB condition and hemorrhage severity, which might explain the discoloration on fillet surfaces. Other authors hypothesized that connective tissue, white striations, excessive moisture and the yellowish fluid on the surface of the fillets can also contribute to the discoloration (Sihvo et al., 2014; Dalle Zotte et al., 2017). The discoloration present in the cooked WB fillets may cause consumer rejection.

1.5.2.3. Instrumental texture profile and Sensory Analysis

Texture is one of the most important attributes used by consumer to perceive quality. Several studies showed that WB is not a problem associated with tenderness considering that no differences in Meullenet-Owens razor shear total energy (MORSE) and Warner-Bratzler shear force, sarcomere length, and gravimetric fragmentation index (GFI) have been found (Mudalal et al., 2014; Tasoneiro et al., 2016; Tijare et al., 2016).

Within this context, Mudalal et al. (2014) reported that raw WB fillets, regardless of the presence of WS, exhibited higher instrumental hardness than normal samples. Tasoneiro et al. (2016) developed a descriptive sensory profile of non-marinated WS and WS/WB fillets, and found that WS/WB samples were tougher compared to the normal and WS ones. In addition, no differences were observed in off-odor, off-flavor, taste and the rest of texture attributes. Furthermore, Aguirre et al. (2018) developed a descriptive texture profile of injection marinated severe WB using two different cooking methods and observed that regardless of the cooking method, nine (springiness, hardness, denseness, cohesiveness, cohesiveness of mass, crunchiness, fracturability, fibrousness and chewiness) out of eleven attributes were significantly higher for WB than normal fillets. The same author found similar results in the instrumental texture profile of severe WB, hardness, springiness, cohesiveness and chewiness were higher compared to normal fillets. Likewise, Soglia et al. (2015) evaluated the instrumental texture traits of non-marinated WB and observed differences in hardness, gumminess and chewiness. On the other hand, Solo (2016) evaluated the consumer perception towards WB meat, results indicated that consumers had lower acceptability of severe WB than normal breast in terms of overall impression and texture. In addition, the open-ended comments revealed that participants had more dislike comments related to texture than like comments. Overall, the texture profile of WB with or without WS is different compared to normal.

1.5.2.4. Histology and Physicochemical quality

White striping and woody breast are recent conditions mainly affecting the *pectoralis major* muscle, although the etiology is still not clear, both conditions seem to

be associated with rapid growth rate, large size, and high breast yield (Kuttappan et al., 2012b; 2013b; Lorenzi et al., 2014). According to Kuttappan et al. (2013b), the severity of the myopathic lesions increased as the degree of white striping increased from normal to severe, also *pectoralis. major* muscles exhibited the highest levels of myodegeneration compared to *pectoralis minor* muscles. Within this context, Kuttappan et al. (2017) reported that fillets exhibiting severe WS/WB individually or together had higher lesion scores when compared to normal regardless of the age (6 to 9 week). Furthermore, the severity of gross and histological lesions was more prominent in fillets affected by the WB condition than fillets affected by WS. In fact, the highest amount of fibrosis was usually detectable in samples affected by WB compared with those affected by WS (Kuttappan et al. 2013b; Mazzoni et al., 2015; Sihvo et al., 2014). In addition, they show similar histological lesions. Soglia et al., 2015 evaluated the impact of WB with or without WS on muscle histology and chemical composition and found that WB fillets showed abnormal rounded fibers with different cross-sectional area and nuclei. Similar results were reported by Sihvo et al. (2014). In addition, fillets affected by the lesions had different amounts of connective tissue and fibrosis within muscle fibers, and increased fat content. Some fibers presented degeneration and inflammation together with chronically damaged muscle. Moreover, observations showed that muscle fibers were replaced with connective tissue, which explains the higher fat content. As a consequence, fillets affected by this condition showed a complete reorganization of skeletal muscle structure (Kuttappan et al., 2013a; Sihvo et al., 2014; Soglia et al., 2015).

The chemical composition of the breast muscle was also affected by both myopathies. Proteomic profile results revealed that WS/WB fillets had higher iron, sodium, and lower potassium, phosphorous and ash compared to the normal ones. Fillets containing WB had more calcium compared to normal and WS/WB (Soglia et al., 2015; Tasoneiro et al., 2016). On the other hand, Tasoneiro et al. (2016), and Petracci et al. (2014); Soglia et al. (2015) reported that WS and WB fillets had increased moisture, fat and collagen together with lower protein levels.

The excess of calcium builds up in WB fillets can be considered as a trigger of necrosis in myofibers since they store higher levels of calcium in the sarcoplasmic reticulum. In fact, as a result of the extracellular calcium influx and the release of the intracellular stores of calcium from the damaged sarcolemma structure and sarcoplasmic reticulum, myofibers can undergo calcium-induced necrosis (Zachary and McGavin, 2012). Indeed, McLennan (2000) reported that prolonged, high intracellular calcium concentrations ($>10 \mu\text{M}$) exert a detrimental effect on cells and induce apoptosis (death of cells). Likewise, the increased amounts of Na in WB and WSWB muscles can lead to higher Ca uptake leading to muscle damage. In addition, the increased levels of sodium and calcium might activate phospholipase, which is a specific enzyme involved in membrane damage (Sandercock and Mitchell, 2004; Zambonelli et al., 2017). Thus, explaining the histological lesions such as muscle degeneration and necrosis in WB muscles.

Soglia et al. (2015, 2016) measured water mobility by the use of low field nuclear magnetic resonance (NMR) and reported that WS and or WB tissue contains a

higher proportion of extra-myofibrillar water, which overall was more loosely bound than normal breast muscle. This way explains the higher moisture content and lower water holding capacity in affected muscles specially WB. According to Sihvo et al. (2014) the increase in moisture content may be explained by the occurrence of moderate to severe edema (fluid accumulation) as a result of inflammatory process in fillets affected by the WB condition. As for fat content, severe cases in both conditions can have up to 2% more fat compared to normal fillets, research studies in sensory analysis found that this amount in fat content might change the flavor profile of both woody and white striping chicken breast (Soglia et al., 2015; Kuttappan et al., 2012a). Although, WS is similar to marbling in appearance, it is not the same. Marbling is associated with increased deposition of fat in the deeper layer of red muscles and it is not associated to any disorder or disease (Harper and Pethick, 2004), whereas, WS apparently replaces damaged fibers and its mainly located in the outer layer of white muscles (Soglia et al., 2015). Furthermore, previous research observed that the changes of the skeletal muscle structure due to muscle degeneration resulting in replacement of muscle fiber with fat and connective tissue caused a decreased protein content. One of the problems with the decreased protein content can be the reduction of actin and myosin, which are responsible for WHC. In fact, Soglia et al. (2015) reported that the incidence of WB and WS/WB had a significant effect on sarcoplasmic and myofibrillar proteins. Both WB and WS/WB conditions had a lower abundance of LC1 slow-twitch myosin light chain and higher relative abundance of 30 kDa troponin T fragment, and 70 kDa myosin heavy chain fragment myofibrillar proteins, with respect to normal fillets. As for

sarcoplasmic proteins, results revealed that most of the enzymes involved in glycolytic-gluconeogenesis pathways are different in fillets affected by WB and WS/WB than the normal ones. Affected samples showed similar and increased amounts of lactate dehydrogenase, glyceraldehyde dehydrogenase, and aldose. Whereas, glycogen phosphorylase and phosphoglucose isomerase were significantly lower. Moreover, WB and WS/WB samples had lower pyruvate kinase and creatine kinase levels. Previous studies have shown that lower levels of creatine kinase are the result of the differences in contractile and metabolic functions due to the increased calcium concentration that induces cellular breakdown with the subsequent loss in intracellular constituents (Mitchell, 1999; Sandercock and Mitchell, 2004). Therefore, explaining the degradation in affected muscles and their impairment effect in overall meat quality.

1.5.3. Hypothesis on the mechanisms determining the onset of WS and WB

Several studies have been performed to try to understand what is causing these two myopathies but the exact reason is still unknown. However, there are different hypotheses about the etiology of these conditions such as changes in satellite cells, fiber structure, and oxidative stress (Sihvo et al., 2014; Soglia et al., 2016; Mazzoni et al., 2015; Mutryn et al., 2015; Zambonelli et al., 2017).

One of the characteristics of WS and WB is the larger muscle fiber diameter without significant changes in muscle fiber number compared to normal breast (Dransfield and Sosnicki, 1999). The problem with it is that there is a reduction in the available space to deliver nutrients, inadequate oxygen supply and removal of waste

products such as lactic acid. Therefore, causing hypoxia (a lower oxygen concentration in affected muscle fibers), oxidative stress (there is an increase in free radicals and there are not enough antioxidants to detoxify them), damages to the muscle fibers and necrosis (Wilson et al., 1990; Sihvo et al., 2014; Zambonelli et al., 2017). In particular, Zambonelli et al. (2017) observed that samples containing WS and WB presented higher levels of crystallin alpha B, adenosine deaminase, MB genes and decreased activity of reactive oxygen species modulator 1, which are involved in the response to reactive oxygen species. In the same line, Mutryn et al. (2015) reported that excessive accumulation of reactive oxygen species within the muscle tissue of affected samples might be involved in initiating the inflammatory mechanism associated with both abnormalities. Moreover, without sufficient circulatory supply, satellite cells mediated repair and regeneration of muscle fibers are suppressed. When the satellite cells cannot repair damaged myofibers, fibrosis takes place, with connective tissue replacing muscle fibers, which affects the contractile properties of the muscle (Dransfield and Sosnicki, 1999). Overall, research suggests that selection for fast growing and high breast yield broilers are responsible for the changes in the skeletal muscle of breast fillets.

1.5.4. Current strategies to alleviate the incidence and severity of WS and WB

Given the increasing incidence of WS and WB abnormalities and considering the high economic losses due to meat quality changes and consumers complains, the poultry industry is currently investigating possible alternatives to mitigate the problem.

Although so far no dietary supplements or nutritional strategies seem to reduce the incidence of WS and WB in the field, some research reported some nutritional strategies

to reduce the severity of these conditions. Kuttappan et al. (2012b) found that a low-fat diet can decrease both the growth rate and fillet weight in birds, resulting in a decrease in the percentage of fillets with severe WS compared with birds fed a high-fat diet. In addition, Radaelli et al. (2017) observed less muscle damage in broilers with feed restricted diets compared to the broilers fed ad libitum. Furthermore, the inclusion of arginine, vitamin C, or reduced amino acid density in the diet has shown a decrease in severity of WB. Arginine improved vasculature in affected muscle fibers, vitamin C reduced oxidative stress, and reduced amino acid density had an impact on satellite cells during the recovery phase (Bodle et al., 2018). Another research study in chicken nuggets suggested that 30% of WB meat could be included in the formulation without making major changes in the texture profile and overall perception of the product (Qin, 2013).

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2. CITRUS FIBER AND RICE STARCH MARINADE INGREDIENTS' EFFECT ON THE QUALITY TRAITS AND TEXTURE PROFILE ANALYSIS OF WOODY BREAST MEAT

2.1. Abstract

The effects of phosphates on the quality traits and texture evaluation in woody breast (WB) meat have been previously study. However, there is limited information on the use of alternative marinade ingredients. Thus, the objective of this experiment was to evaluate the effect of citrus fiber (CF) and rice starch (RS) on the quality traits and texture profile analysis of WB meat. A total of n=400 normal (NOR) and severe WB were evaluated. Drip loss, color and pH were measured at 24h postmortem. Treatments; Control) NaCl (0.83%)+STPP(0.45%), T1) NaCl(0.83%)+STPP(0.45%)+RS(1.5%), T2) CF(1.3%), and T3) CF(1.3%)+STPP(0.45%) were evaluated. Individual fillets were injected with 18% brine and 20min, 2h and 24h retention was determined. At 24h post-marination, color and pH were determined and then fillets were cooked (73°C) in foil-covered pans in a convection oven (177°C) and cook loss was determined. At 24h post-cooking, texture profile analysis was performed. Texture samples were cut into 3 rectangular pieces (4x2cm). Non-marinated WB fillets exhibited higher drip loss, L*, b*, and pH compared to normal fillets ($P \leq 0.05$). For RS, regardless of the treatment WB had lower pick up yield (PUY), and higher color and pH than normal. No differences were found between WB RS+STPP and NOR STPP at 20min, 2h and 24h retention; however, WB STPP had the lowest retention values. Cook loss was no different between RS+STPP NOR and WB. WB had different texture profile than NOR fillets. For CF

(Control, T2, T3), regardless of the treatment, WB had lower PUY and 24h retention, and higher a^* and b^* values compared to NOR ($P \leq 0.05$). T3 showed higher 24h retention values compared to the control and T2. No differences in cook loss were found among NOR control, T2 and WB T2. Cohesiveness was higher in WB when compared to NOR. No differences in texture were found between T3 NOR and WB. The results of this study suggest that rice starch and citrus fiber can improve meat quality in severe WB fillets. Citrus fiber is a clean label ingredient that can be used as STPP substitute in marinade brines.

2.2. Introduction

Woody breast (**WB**) is a condition which negatively affects the *pectoralis major* muscle. It is characterized by abnormal hardness with different degrees of severity. The resulting texture of this WB meat is described as crunchy and fibrous. Morphological variations associated with WB meat are pale color, hemorrhaging, and exudate on the surface of the fillets (Sihvo et al., 2014; Kuttappan et al., 2016, 2017; Aguirre et al., 2018). In addition, the more severe WB meat can be associated with greater depth of the *pectoralis major* in the cranial and medial regions of the fillet. According to Sihvo et al. (2017), WB is a polyphasic myodegeneration in broilers and its morphological changes are detected as early as 2 weeks of age. Woody breast has been regularly found in fast-growing and high-yielding strains; thus, the incidence and severity increase as the birds become older and heavier (Kuttappan et al., 2017). Recent publications also reported a significant reduction of functional and total protein content and higher moisture and lipid levels in WB (with or without white striping) compared to normal fillets (Soglia et al., 2016a, b; Baldi et al., 2018). Moreover, meat affected by the WB condition shows reduced protein functionality resulting in higher drip loss, and reduced ability to hold water in both raw and cooked meats (Mudalal et al., 2015; Soglia et al., 2016a; Dalgaard et al., 2018; Baldi et al., 2018).

Marination has been widely used by the poultry meat industry to improve flavor, tenderness, and protein functionality (Alvarado and Sams, 2003; Alvarado and McKee, 2007). Previous research has shown that the use of sodium chloride (**NaCl**) and sodium tripolyphosphate (**STPP**) increase the number of negative charges within the meat

structure and therefore, increases water holding capacity (**WHC**) in normal (**NOR**) and PSE meat (Woelfel and Sams, 2001). Within this context, a number of studies have shown that WB meat marinated with NaCl and STPP showed lower marinade uptake, retention, and higher cook loss % regardless of the meat pH, marination method (injection or tumbling), or cooking method (Mudalal et al., 2015; Tijare et al., 2016; Bowker et al., 2018; Aguirre et al., 2018).

Over the last few years, consumers have decreased their support of phosphates as a marinade ingredient because of nutritional concerns related to lower absorption of minerals inside the intestinal track (Sherman and Metha, 2009). In addition, the term “phosphate” sometimes has negative connotations (Petracci et al., 2013). Therefore, alternative marinades have become more popular in retail cuts.

The use of organic marinade ingredients such as rice starch and citrus fiber has been increasing within the meat industry because they provide similar characteristics compared to phosphates. Remyline AX DR (Beneo, Belgium) is a clean-label, waxy rice starch used to improve the cook yield and overall final yield of tumbled or injected chicken breast. Likewise, Elite Start Citrus fiber (Formtech Solutions Inc., Schenevus, NY) is a clean label ingredient mainly composed of citrus flour and natural flavorings. This citrus fiber is often used to improve the water holding capacity in marinated raw poultry products.

Considering that there is limited information in the use of alternative marinade functional ingredients in the performance of the quality of WB, the objective of the study

was to evaluate the effect of rice starch and citrus fiber marinade ingredients on the quality traits and texture profile analysis of severe WB meat. The hypothesis of the study was that rice starch and citrus fiber will improve meat quality and the texture attributes of fillets affected by the woody breast condition.

2.3. Materials and Methods

2.3.1. Meat preparation

Normal and severe WB fillets were obtained 3 h postmortem (**PM**) from a commercial facility and transported to the Poultry Science Research Center at Texas A&M University. The meat samples were categorized into NOR (0) and severe WB (3) by tactile evaluation of hardness following the classification proposed by Tijare et al. (2016). Over two separate trial runs, a total of 200 NOR and 200 severe WB breast fillets were used in the study. Upon arrival at the facility, the breast fillets were trimmed to remove the excess of fat and cartilage. At approximately 5 h PM individual fillets were weighed and placed in storage Ziploc® bags for the determination of drip loss.

2.3.1.1. Pre-injection

After 24 h PM, drip loss (%), color (*L=lightness, *a=redness, *b=yellowness), and pH were measured from each fillet. Color was measured from the bone side of each fillet using the CIELAB *L = lightness, a* = redness, and b* = yellowness color scale of a calibrated colorimeter (Chroma Meter Measuring Head CR-400; Konica Minolta, INC., Japan). The pH was measured from the cranial end of each fillet using a pH meter (Piercing probe Model 205; Testo, Inc., Sparta, NJ).

2.3.1.2. Post-injection

Right after quality measurements were taken, two hundred NOR and two hundred severe WB fillets were randomly assigned to four ($n = 50$ each) different treatments. The meat was marinated using a multi-needle injector (Inject-star BI-88 P-VSO, Mountain View, AZ) to 18% (green weight) target at constant pressure (15 to 20 psi). Four ingredients were used in different combinations: NaCl, RS (Remyline AX DR, Beneo, Belgium), STPP (Gusto M31, Formtech Solutions Inc., College Station, TX) and CF (Elite Start, Formtech Solutions Inc., Schenevus, NY). The treatments consisted of the following commercial formulations based on the final concentration in the meat:

1. Control - NaCl (0.83%) + STPP (0.45%)
2. Trt 1- NaCl (0.83 %) + STPP (0.45%) + RS (1.5%)
3. Trt 2- CF (1.3%)
4. Trt 3- CF (1.3%) + STPP (0.45%)

Fillets were weighed immediately, 20 min, 2 h and 24 h for the determination of marinade pick up and marinade retention. After 24 h post-marination, color ($*L$, a^* , and b^*), and pH were measured from each fillet as explained previously. Then, fillets were cooked to an endpoint internal temperature of 73°C in a convection oven (FC-34/1 Sodir Convection Oven, Equipex, Providence, RI) set at 177°C , using pans covered with aluminum foil. During cooking, internal temperatures were monitored using copper-constant thermocouples (Omega Engineering, Stamford, CT) inserted into the geometric center of the cranial end of the fillets. Cooked samples were allowed to cool down for 15

min and reweighed to calculate cook loss. Then, fillets were placed at refrigeration temperatures (3° C) overnight until instrumental evaluation.

2.3.2. Texture Profile Analysis (TPA)

The ventral cranial end portion of the fillet was cut into 3 rectangular 4 cm × 2 cm samples using a template and a sharp knife (Mudalal et al., 2014, Aguirre et al., 2018). The texture measurements were performed using a texture analyzer (TA.XTPlus, Texture Technologies, Hamilton, MA) with a cylinder probe of 76.2 × 10 mm to compress the samples. A 50 kg load cell at a pre-test speed of 3.0 mm/sec, 1 mm/sec test speed, 3.0 mm/sec post-test speed and 2.0 sec in between compressions was used to reach a 25% compression of its initial height.

2.3.3. Statistical Analysis

Considering the nature of both ingredients where rice starch is used to improve meat quality under hot conditions (during cooking) and citrus fiber is used during cold conditions (processing and shelf life of raw products), the analyses was performed separately. Phosphate was used as a control in the analysis of both ingredients. Data was analyzed by ANOVA using the least squares procedure of JMP Pro (version 14.0 SAS Institute Inc.). For the non-marinated data drip loss (%), color and pH, the model included type (NOR and SWB) as fixed effect and replication as a random effect. For the marinated data pick up yield (PUY), color and pH, the model included type and treatment as well as their interactions as fixed effects and replication as random effect. Pick up yield was used as a covariate to control the potential effect on marinade

retention, cook loss and TPA. The experimental unit was individual fillets. Means were separated by Tukey's HSD test at a significance $P \leq 0.05$.

2.4. Results and Discussion

The experiment was performed to evaluate the effect of citrus fiber and rice starch marinade ingredients on the quality traits and texture profile analysis of severe WB meat.

The results for non-marinated meat quality values (24 h PM) are shown in Table 1. As expected, WB fillets displayed higher drip loss % ($P = 0.001$), lightness ($P = 0.007$), yellowness ($P = 0.02$), and pH ($P = < 0.0001$) compared to NOR breast. Previous research has indicated similar results (Mudalal et al., 2015; Chatterjee et al., 2016; Kuttappan et al., 2017; Aguirre et al., 2018; Bowker et al., 2018). Normally, water in meat can be considered bound (between myofilaments), immobilized (between myofibrils) or free (outside the fibers). In this regard, Soglia et al. (2015, 2016) measured water mobility by the use of low field nuclear magnetic resonance (NMR) and reported that WS and or WB tissue contains a higher proportion of extra-myofibrillar water, which overall was more loosely bound than normal breast muscle, which can further explain the higher drip loss found in WB compared to NOR.

The marinated meat quality parameters for rice starch are reported in Table 2. There was no interaction between meat type and treatment ($P \geq 0.05$). Fillets with the WB condition had lower PUY percentage ($P = < 0.0001$), higher L^* ($P = < 0.0001$), a^* ($P = 0.0005$) and b^* ($P = 0.02$) values, and higher pH ($P = < 0.0001$) compared to NOR.

Mudalal et al. (2015), Tijare et al. (2016), Bowker et al. (2018), Aguirre et al. (2018) reported similar results in WB marinated with salt and phosphate. Regardless of the meat type, RS + STPP had approximately 2.29 % less PUY ($P = <0.0001$), higher color values ($P = < 0.0001$), and lower pH ($P = 0.0008$) than the STPP treatment. These results suggested that the raw meat color and pH of WB are not affected by the addition of RC + STPP.

In regards to the marinade retention and cook loss for rice starch (Table 3), there was a significant interaction between the type and treatment ($P \leq 0.05$). Woody breast fillets injected with RS + STPP had similar 20 min, 2 h and 24 h post marinade retention compared to NOR STPP. In addition, severe WB fillets injected with RS + STPP showed higher overall marinade retention compared to WB STPP ($P \leq 0.05$). As for cook loss, NOR (13.04 %) and WB (14.64 %) injected with RS + STPP treatment were not significantly different and had the lowest cook loss % compared to NOR (24.83 %) and WB (33.40 %) injected with STPP ($P = < 0.0001$). WB injected with the STPP showed the highest cook loss compared to the variables ($P = < 0.0001$). These results suggest that the inclusion of RS + STPP in the brine affect the cook loss of NOR and WB in a similar fashion, thus, reducing the cook loss by more than 18% compared to WB injected with STPP alone. Phosphates aid in the development of a protein structure network by increasing the electrostatic repulsive forces which expand the spaces between myofibrillar proteins (actin and myosin) allowing for more water to be entrapped in these gaps (Barbut, 2002). On the other hand, starch absorbs free water and forms a gel during the cooking process (Petracci et al., 2013). Therefore, both

ingredients could be considered to have complementary effects (Resconi et al., 2015). This observation agrees with the finding described by Resconi et al. (2015), who performed NMR (nuclear magnetic resonance) analysis in whole muscles hams injected with rice starch and STPP and found that during the cooking process rice starch retained free water due to the rice starch gel formation. Considering that WB meat has high amounts of free water (Baldi et al., 2019), this may explain the significant differences in cook loss between WB RS + STPP, and WB STPP.

The marinated meat quality parameters for citrus fiber is reported in Table 4 and 5. No interaction was found between type and treatment for PUY, 24 h retention, and a^* and b^* values ($P \geq 0.05$). As expected, fillets with the WB condition showed lower PUY percentage ($P = < 0.0001$), lower 24 h post marination retention ($P - \text{value} < 0.0001$), and higher a^* values ($P = < 0.0001$) compared to NOR. No differences were observed in yellowness ($P = 0.2395$). As for the treatments, regardless of type of meat, CF had higher PUY (%) compared to CF + STPP, and the STPP treatment ($P = 0.0001$). After 24 h post marination, CF + STPP had a higher retention than CF, and STPP ($P = < 0.0001$). No differences were found in redness ($P = 0.0585$), but yellowness values were higher for CF + STPP, and CF than STPP ($P = < 0.0001$).

There was an interaction between type and treatment for 20 min and 2 h retention, L^* , pH, and cook loss ($P \leq 0.05$). As shown in Table 5, NOR CF + phosphate, and NOR CF had a higher 20 min retention compared to the rest of the treatments ($P = 0.0003$). After 2 h post marination, no differences were found between WB CF+STPP, NOR STPP, and NOR CF ($P \geq 0.05$). In addition, NOR CF + STPP had higher 2 h

retention than the rest of the variables ($P = 0.0005$). Moreover, WB CF + STPP, WB CF, WB STPP, and NOR CF + phosphate were lighter than NOR CF and NOR STPP (P – value 0.0017). In regards to the pH, WB CF + STPP, WB STPP, and NOR CF + STPP had higher values compared to the rest of the variables ($P = 0.0082$). These results are in agreement with prior research in normal breast fillets where Casco et al. (2013) reported no differences in marinade retention (2 h and 24 h), higher b^* values and higher pH when comparing a phosphate blend and a citrus fiber blend (Savorphos A-F200). Fernández-López et al. (2004), Aleson-Carbonell et al. (2003), Aleson-Carbonell et al. (2004) also found an increase in b^* values and concluded that presence of yellow components in the citrus fiber (albedo). As for cook loss (Table 5), WB STPP had the highest cook loss followed by WB CF + STPP, and WB CF which is expected as CF is meant to bind and retain water in a raw system during processing and shelf-life as opposed to rice starch which is a better binder (swells with water) in cook systems to increase cook yield (Petracci et al., 2013). Considering the poor quality of severe WB, it was interestingly to find that WB CF (26.56 %) compared as well as the NOR CF (24.22 %) and NOR STPP (23.74) in cook loss. Normal CF + STPP had the lowest cook loss compared to the other treatments ($P = < 0.0001$). Similar results were reported by Aleson-Carbonell et al. (2005), who found that the use of citrus albedo improved the cooking properties of beef patties. Likewise, Casco et al. (2013) reported improved cook loss in breast fillets injected with citrus fiber.

Even though no significant differences in cook loss were found between WB CF + STPP, and WB STPP, there was a 3 % cook loss numerical reduction in WB fillets

injected with CF + STPP, which can make a yield difference for industry. In addition, the improved cook loss in NOR fillets injected with CF + STPP could improve consumer acceptance of retail and restaurant cuts. These results suggest that CF or CF + STPP can improve the final yield in WB meat. In addition, CF can be a potential clean label ingredient substitute for STPP in marinade brines, which can improve consumer perception towards poultry products.

2.4.1. Texture Profile Analysis (TPA)

Texture profile analysis is a double compression test that mimics the mouth's biting action and it is often used for the determination of the textural properties of foods (Texture Technologies, 2019). The primary parameters evaluated in chicken breast are: hardness, which is described as the peak force that occurs during the first compression; springiness, is how well the product physically springs back after it has been deformed during the first compression and has been allowed to wait for the target wait time between strokes; cohesiveness is how well the product withstands a second deformation relative to its resistance under the first deformation; and chewiness is defined as the energy required to masticate a solid food to a state ready for swallowing and it is measured as a product of hardness, cohesiveness, and springiness (Muñoz et al., 1992; Texture Technologies, 2019).

Texture parameters for rice starch are reported in Table 6. There was no interaction between type of meat and treatment ($P = 0.6674$). Woody breast fillets had higher hardness, springiness, cohesiveness and chewiness compared to NOR fillets ($P \leq$

0.05). Only cohesiveness was slightly higher in RS +STPP compared to STPP ($P = 0.0281$). The increased cohesiveness can be the result of the rice starch gel formation which improved the stability of breast fillets. Similar results were found by Resconi et al. (2015). Data suggest that there are not significant changes in the overall texture profile of breast fillets, therefore, RS + STPP can be used as an alternative to mitigate the poor meat quality in severe WB fillets.

Texture parameters for citrus fiber are reported in Table 7. No interaction was found between type and treatment for cohesiveness ($P = 0.6344$). Woody breast fillets were slightly more cohesive compared to NOR ($P = 0.0006$). Aleson-Carbonell et al. (2005), investigated different types of citrus fiber (lemon albedo) in the characteristics of beef burger and found that cohesiveness was not affected by the treatment. In regards to the treatments, CF + STPP was slightly more cohesive than STPP, and CF ($P - \text{value} < 0.0001$). Texture attributes hardness, springiness and chewiness presented an interaction between type and treatment ($P \leq 0.05$). Interestingly, the texture attributes were similar for WB CT + STPP and NOR CF + STPP. Moreover, no differences were found in chewiness between WB CF, WB CF + STPP, and NOR CF + STPP. In this regard, Casco et al. (2013) evaluated the consumer perception of breast fillets injected with citrus fiber and phosphate using a triangle test and found that consumers were not able to detect differences between the two products. Overall, results suggest that the inclusion of CF and CF + STPP in the marination brine can improve the texture of WB and actually help to make the WB meat feel more like normal meat. Further research with consumers is need it to confirm these results.

2.5. Conclusions

Results from this study indicate that rice starch + STPP, citrus fiber, or citrus fiber + STPP could be used to improve marinade retention and cook loss yield in breast meat affected with the WB condition. With the rising demand for clean label ingredients, citrus fiber could be a potential clean label ingredient used as substitute of STPP in marinade brines. Sensory analysis should be performed to investigate the overall consumer perception towards the inclusion of this ingredients in the final product.

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3. FRESH SAUSAGE AND DELI-LOAVES CHICKEN PRODUCTS FORMULATED WITH DIFFERENT PERCENTAGES OF WOODY BREAST MEAT

3.1. Abstract

Quality traits and texture evaluation of woody breast (WB) as a whole muscle have been previously assessed. However, there is limited information about the inclusion of WB meat in processed products without altering quality and texture. Therefore, two experiments were conducted to determine the effects on quality traits, acceptability, and instrumental texture of fresh chicken sausage (Exp. 1) and deli-loaves (Exp. 2) formulated with severe WB and/or normal (NOR) meat as follow: T1) 100% NOR+0% WB, T2) 75% NOR+25% WB, T3) 50% NOR+50% WB, T4) 25% NOR+75% WB, and T5) 100% NOR+100% WB. For both experiments, WB and NOR meat were obtained from a commercial facility (< 24h postmortem). Exp. 1, color (L^* , a^* , and b^*), pH, proximate analysis (fat and moisture), cook loss, texture profile analysis (TPA), and consumer sensory were measured. No differences in color were detected among the treatments for both raw and cooked sausage ($P \leq 0.05$). pH gradually increased as the percentage of WB increased. T1 had the lowest moisture content compared to the rest of the treatments. For cook loss, there was no difference between T1 WB (11.42; 10.13) and T5 WB ($P \leq 0.05$). However, T4 WB (7.61) had the lowest value ($P \leq 0.05$). No differences in hardness and chewiness were found between T1, T2, T3 and T5 ($P \leq 0.05$). Consumers were only able to detect texture differences between T1 and T5 ($P \leq 0.05$). This experiment suggested that WB could be included in sausage formulations up to

75% without affecting the quality traits and consumer perception. Exp. 2, color, pH, cook loss and TPA were evaluated. L*, b*, and pH values gradually increased and a* values decreased with the incorporation of WB in the formulation. T5 had the highest cook loss among the treatments ($P \leq 0.05$). No differences in hardness and chewiness were found. However, differences in resilience, cohesion and springiness were detected between the treatments ($P \leq 0.05$). T4 The addition of up to 75% WB (T4) could be a suitable option in the formulation of deli-loaves without affecting the texture and yield of the final product in both deli loaves and sausages.

3.2. Introduction

Among the problems affecting the economic value of broiler breast muscle is the impaired quality of woody breast meat (**WB**). For the US broiler market, Kuttappan et al. (2016) roughly estimated an economic loss of \$200 million per year due to this condition. Woody breast is a modern condition affecting the *pectoralis major* muscle. It is characterized by abnormal hardness with different degrees of severity and poor meat quality properties related to decreased functional protein, increased collagen, and increased fat. Fillets with the WB condition also exhibit a higher pH compared to NOR fillets due to a decrease in the glycolytic potential and altered energetic status and metabolic pathways found in previous studies resulting in a higher ultimate pH after rigor mortis (Zambonelli et al., 2016). However, although the ultimate pH of fillets affected by either WB and WS or both conditions in the same fillets is higher compared to normal, their WHC is significantly lower in non-marinated and marinated fillets (Soglia et al., 2016a; Tijare et al., 2016; Bowker et al., 2018). Woody breast condition also affects the texture and appearance (associated with fillets discoloration), and therefore, consumer willingness to buy raw and cook fillets (Mudalal et al., 2015; Chatterjee et al., 2016; Kuttappan et al., 2017; Aguirre et al., 2018; Zhuang and Bowker, 2018).

Other studies have investigated alternative processing solutions to mitigate the effect of WB in breast fillets. Sanchez-Brambila et al. (2017) investigated the quality properties and the texture profile of cooked patties made of ground WB fillets and found that WB patties are less springy and chewy and presented no differences in cook loss

compared to normal (**NOR**) patties. Similarly, Chen et al. (2017), evaluated the functional properties of meatballs made of WB, salt, and tripolyphosphate and found that WB meat is not suitable to produce gel-type products because it affects the texture and functional properties of the final product. Considering that very few studies have investigated the inclusion of different percentages of WB in processed products and that there is scarce information about final product quality related to the inclusion of WB in sausage and deli-loaves. Two experiments were conducted to determine the effects on quality traits, acceptability, and instrumental texture of fresh chicken sausage and deli-loaves formulated with 0%, 25%, 50%, 75% and 100 % severe WB meat. The hypothesis of the study was that chicken sausage and deli-loaves containing up to 100% WB will not impact meat quality and texture profile.

3.3. Materials and Methods

3.3.1. Sample Collection and Preparation

For the two experiments, boneless skinless NOR and severe WB fillets were obtained 3 h postmortem from a commercial facility and transported to the Poultry Science Research Center at Texas A&M University. The meat samples were categorized into NOR (0 = flexible throughout the fillet) and severe WB (3 = very hard throughout the fillet) by tactile evaluation of hardness following the classification proposed by Tijare et al. (2016). Then, breast fillets were trimmed to remove the excess of fat and cartilage.

3.3.2. Experiment 1-Fresh Chicken Sausage

3.3.2.1. Sample preparation

Fresh chicken sausage was made using a commercial formulation composed of pork trimmings (50 % fat, 50% meat), chicken breast meat (NOR and/or WB), 6% water, 3% spices (Mild Italian Sausage Seasoning, Old Plantation Seasonings, A.C. Legg, Inc.) and 0.42 % phosphate. The treatments consisted of the following concentration of WB in the formulation:

1. 100% NOR + 0% WB
2. 75% NOR + 25% WB
3. 50% NOR + 50% WB
4. 25% NOR + 75% WB
5. 0% NOR + 100 % WB

After the meat/nonmeat ingredients were weighed, chicken breast and pork trimmings were ground (Meat grinder #22, Avantco® Equipment) separately through a 3.81 cm grinding plate, and then were formulated to contain 75% lean source and 25% fat source. Once formulated, they were mixed for one min in a paddle mixer (Commercial Food Mixer, Texas Tastes, TX). Following mixing, dry ingredients were added and mixed thoroughly for two min. After all ingredients were mixed together, a final grind (0.95 cm grind plate) was performed and five batter samples were collected per each treatment for further meat quality evaluation (color, pH, fat and moisture). Subsequently, the sausage batter was stored at 3°C for 48 h to improve protein extraction

(Hargarten, 2015), and then vacuum-stuffed in collagen casings (26 mm fresh collagen casing, The Sausage Maker Inc, New York). Over three separate trial runs, a total of 75 sausage units were used in the study. Approximately 0.42 kg ($P = 0.09$) was distributed into 6 sausage links, therefore, the group of 6 sausages was considered as 1 experimental unit. The sausages were placed on metal racks with aluminum pans and cooked to 73°C internal temperature using a convection oven (FC-34/1 Sodir Convection Oven, Equipex, Providence, RI) set at 177 °C. During cooking, internal temperatures were monitored using copper-constant thermocouples (Omega Engineering, Stamford, CT) inserted into the center of the sausages. After the product was cooked to the final product temperature, they were removed from the oven and cooled for 15 min. The sausages were weighed for cook loss determination, packaged in Cryovac vacuum-package bags (Model B4173T, Sealed Air Corporation, Simpsonville, SC), and stored in a blast freezer at -20°C.

3.3.2.2. Meat quality measurements and proximate analysis

Color measurements (L^* =lightness, a^* =redness, b^* =yellowness) were collected ($n = 6$ per trt) using a 3 reading average of the ground raw meat, sausage batter, and cooked product with a calibrated Chroma Meter Head (CR-400 Head, Konica Minolta, Inc.). pH ($n = 6$ per trt) was measured using a piercing probe (Model 205; Testo, Inc., Sparta, NJ). Proximate analysis ($n = 9$ per trt) was conducted using the AOAC Official Method 960.39 (Soxhlet Extraction of Crude Fat) for the determination of fat and moisture content in each sausage treatment.

3.3.2.3. Sensory Analysis

A consumer sensory analysis (IRB2016-0783M) was performed to evaluate fresh chicken sausage at the Texas A&M Kleberg Center Sensory Kitchen. Sausages were cut in 2.6 cm × 2 cm cylinders, and 2 pieces were served for evaluation in an odorless plastic plates (15.87-cm, Member's Mark, USA). Each sample was assigned a random 3-digit code and samples were randomly assigned to a different serving order. All the samples were tested during 1 d of evaluation. Consumers (n = 80) were randomly selected and placed in individual breadbox-style booths separated from the preparation area under regular lights. Consumers received 5 sausage samples, each containing different percentages of WB (0 %, 25%, 50%, 75%, and 100%); double-distilled deionized water and unsalted crackers for palate cleansing between samples; the demographic sheet; and a sensory ballot. Overall, color, flavor, texture and juiciness were included on the ballot using 9-point hedonic scales (0 = dislike extremely, 9 = like extremely) as well as two open-ended questions asking what they liked or disliked about the samples.

Prior to analysis, sausages were thawed at refrigeration temperatures (3° C) overnight. Sausages were re-heated (177 °C oven temperature) on aluminum pans to an average of 73 °C internal temperature. Then they were placed in a holding oven at 48.8° C on a plate covered in aluminum foil for no more than 20 min or served immediately in order to not affect sensory properties (American Meat Science Association, 2016).

3.3.2.4. Texture Profile Analysis (TPA)

Prior to analysis, sausages were thawed at refrigeration temperatures (3°C) overnight. Subsequently, sausages (n = 9 per trt) were cut into 3 (2.6 cm × 2 cm) cylindrical samples using a template and a sharp knife (Mudalal et al., 2014, Aguirre et al., 2018). The texture measurements were performed using a texture analyzer (TA.XTPlus, Texture Technologies, Hamilton, MA) with a cylinder probe of 76.2 × 10 mm to compress the samples. A 50 kg load cell at a pre-test speed of 3.0 mm/sec, 1 mm/sec test speed, 3.0 mm/sec post-test speed and 2.0 sec in between compressions was used to reach a 30% compression of its initial height.

3.3.3. Experiment 2-Deli-Loaves

3.3.3.1. Sample preparation

Deli-loaves were made using a commercial formulation composed of chicken breast meat (NOR and/or WB), water (18.5 %), salt (1 %) and phosphate (0.5 %). The treatments consisted of the following concentration of WB in the formulation:

1. 100% NOR + 0% WB
2. 75% NOR + 25% WB
3. 50% NOR + 50% WB
4. 25% NOR + 75% WB
5. 0% NOR + 100 % WB

Thirty percent of the meat was ground using a 0.47 cm plate and 70 % of the meat was ground using a kidney plate. Then, a 20% (wt/wt) brine composed of ice

water, salt and phosphate was added into the tumbler (MC-25 Lab Tumbler, Inject Star of the Americas, Inc., Mountain View, AR), a subsequent vacuum was pulled (22 in Hg) and the meat tumbled at 25 RPM for two hours under refrigeration (3°C). Then, the deli-loaves batter was vacuum-stuffed in clear fibrous casings (20.32 cm x 38.1 cm, DeWied International, San Antonio, TX). Over three separate trial runs, a total of 75 deli-loaves were used in the study. The deli-loaves were cooked to 73°C internal temperature in an electric convection oven (Mark V, Blodgett, Burlington, VT) using a loaf cooking program for 4 h. The internal temperature was monitored by copper-constantan thermocouples (Omega Engineering, Stamford, CT) inserted into the geometric center of the loaves. After the product was cooked, it was removed from the oven and cooled for 15 min. Following cooking, deli-loaves were weighed for cook loss determination. They were then packaged in Cryovac vacuum-package bags (Model B4173T, Sealed Air Corporation, Simpsonville, SC) and stored in a blast freezer at -20°C until used for further analysis.

3.3.3.2. Color, pH, and TPA

Prior to analysis, deli-loaves were thawed at refrigeration temperatures (3°C) overnight. Each deli-loaf was sliced into 2 cm thick slices, and two slices from each treatment were used for color and pH, and three slices for TPA measurements. Color (L*=lightness, a*=redness, b*=yellowness) was collected using a 3 reading average of the raw and cooked product with a calibrated Chroma Meter Head (CR-400 Head, Konica Minolta, Inc.) and pH was measured using a piercing probe (Model 205; Testo,

Inc., Sparta, NJ). Texture profile analysis was performed using the same procedure as the fresh sausage.

3.3.4. Statistical analysis

Data were analyzed by ANOVA using JMP Pro (version 14.0 SAS Institute Inc.). For the sausage and deli-loaves, treatments (0, 25, 50, 75, and 100 % WB) were considered as fixed effect and replication as a random effect. Consumer sensory data were analyzed using Kruskal-Wallis Test (Rank Sums) as the data was not normally distributed. Means were separated using Turkey's HSD test at a significance $P \leq 0.05$. Open-ended comments from the consumers were screened to identify recurring terms. The terms having similar meaning were manually grouped into different categories for each treatment. The open-ended comments from the consumers regarding the reason for like or dislike of the chicken sausage were analyzed using The Goodness of Fit Chi Square Test (Version 24, IBM SPSS, Armonk, NY) as proposed by Kuttappan et al. (2012). The association among variables were generated using Pearson correlation coefficients.

3.4. Results and Discussion

3.4.1. Experiment 1 -Fresh Chicken Sausage

Sausages are classified as a ground product made of coarse minced meat where meat fibrous structure is still detectable to some extent (Petracchi et al., 2013). The initial quality of the meat sources used in this study are shown in Table 8. Woody breast meat showed higher pH and color (L^* and b^* values); ($P \leq 0.05$), confirming the effect of the

condition on breast meat. These results were consistent with previous research (Mudalal et al., 2015; Soglia et al., 2016b; Aguirre et al., 2018; Bowker et al., 2018; Zhuang and Bowker, 2018).

The color and pH of the raw (Table 9) and cooked (Table 10) sausage showed no differences in color (L^* , a^* , and b^*) values. However, the pH gradually increased as the percentage of WB increased in both raw and cooked sausages. According to Zotte et al. (2017), breast fillets with greater size could exhibit a reduced glycolytic potential, therefore, explaining the higher pH compared to normal fillets. In addition, Soglia et al. (2016b) hypothesized that the high ultimate pH in breast meat affected by the WB condition is the result of an altered glycogen utilization which leads to glycogen depletion.

Moisture, fat, and cook loss values are shown in Table 11. Regardless of the concentration of WB in the chicken sausage, 0 % WB had the lowest moisture content compared to the rest of the treatments. These results are in agreements with Soglia et al. (2016a), which found that WB fillets had higher moisture, connective tissue, fat and lower protein content compared to normal fillets. Moreover, Chen et al. (2018) reported that the molecular water mobility distribution had different proportions in WB than normal, which explains the higher moisture content in WB incorporated sausage. Fat content was higher for 0% WB compared to 25% and 100% WB, which could be associated to the formulation process, where the pork source could have higher fat to meat ratio. Correlation analysis revealed that cook loss % was not strongly influenced by fat content ($r = 0.35$, $P < 0.0001$) or by pH ($r = - 0.30$, $P < 0.0001$). Therefore, other

factors may play a major roll. For cook loss, no differences were found between 0 and 100 % WB. In this regard, Sanchez Brambila et al. (2017) found no differences in cook loss in patties made with severe WB compared to normal. Moreover, Madruga et al. (2019), reported similar results in emulsion type chicken sausage made with 100% WB meat. Interestingly the incorporation of 25, 50 and 75% severe WB into the sausage mixture significantly reduced the cook loss % ($P \leq 0.05$) compared to the control treatment. Thus, these results may be a reflection of different mechanism of water binding in comminuted products with inclusion of severe WB. Further investigation is needed to determine mechanism of action.

Texture is one of the most important attributes used by consumer to perceive quality. The TPA analysis (Table 12) indicated that 75 % WB was less hard and chewy compared to 0% WB ($P \leq 0.05$). and similar ($P \geq 0.05$) to the rest of the treatments. No differences in cohesion and springiness were found across the treatments ($P \geq 0.05$). Similar results were found by Rigdon et al. (2019), who developed a chicken sausage formulated with abdominal fat and WB meat of varying degrees of severity and found that the values of hardness tended to decrease as the rate of WB increased and no differences were found in cohesiveness and springiness. Likewise, previous studies in whole muscle indicated that the TPA of WB affected fillets were associated to higher hardness, chewiness values (Soglia et al., 2016b; Aguirre et al., 2018). Results in this study demonstrated that when comminuted, the small particles size may negate the hard and chewy texture profile as compared to the whole muscle.

The consumer demographics are shown in table 13. The majority of the consumers were Caucasian (72.50 %) with less than 35 years of age. Gender of the consumers was almost evenly divided. Overall, color, flavor and juiciness were all similar ($P \geq 0.05$, Table 14) across treatments. Consumers did not find differences in texture between treatments 0, 25, 50 and 75 % WB ($P \geq 0.05$). Consumers significantly like the texture of the fresh chicken sausage formulated with 0 % WB, more than the 100%WB ($P \leq 0.05$). Table 15 displays the examples of comments and its frequencies of occurrence based on consumers preference (like or dislike), where only “like appearance”, and “dislike moisture” categories were different ($P \leq 0.05$). Thirty three percent of the consumers explained that they like the appearance of the fresh sausage formulated with 100 % WB because it presents a “good color” or is “less greasy”, whereas 19% like the appearance of 0% WB. These results could be related to the higher fat content in the sausage formulated with 0% WB, however, when consumers were asked if they dislike the appearance (“very greasy”, “little fat”, “pale color”) no differences were found among the treatments ($P = 0.681$) meaning that fat content did not influenced consumer perception. In regards to the “dislike moisture” category, only 9 % of consumers described the sausage formulated with 100 % WB as being “dry” or “very juicy” as opposed to 20 % in the sausage formulated with 0% WB. These results are in agreement with the higher moisture content found in 100% WB compared to 0% WB.

Overall, these results suggest that the comminution process can be an alternative to reduce the impaired texture and quality issues presented in breast fillets with the WB

condition. Moreover, as evidenced by the TPA and then related to consumer sensory panel, severe WB meat could be included in the formulation of chicken sausage up to 75% without affecting the texture perception by consumers.

3.4.2. Experiment 2-Deli-Loaves

Deli-loaves are categorized as a formed/restructured product manufactured by chunks or pieces of meat bonded together (Petracci et al., 2013). Table 16 shows the results of color and pH obtained from raw deli-loaves. L^* values of the 75 and 100% WB deli-loaves were significantly higher than those made by 25 and 0% WB. No differences in lightness were found between treatments 0, 25 and 50% WB ($P \geq 0.05$). Color a^* values decreased as the percentage of WB increased, 0% WB had the highest a^* values among the five treatments ($P \leq 0.05$). For b^* values, 100% WB had higher values compared to 0 and 25% ($P \leq 0.05$). The pH gradually increased as the percentage of WB increased; 75 and 100% presented the highest pH values compared to the rest of the treatments ($P \leq 0.05$).

As for cook deli-loaves (Table 17), L^* values gradually increase as the percentage of WB increased in the formulation, only 25% is similar to 0% WB ($P \geq 0.05$). For color a^* and b^* , 100 % WB had higher b^* values, and lower a^* values compared to the 0% WB. Similar to the raw deli-loaves, pH values were higher for 100% WB than 0% WB. Contrary to our results, Chen et al. (2017) found no differences in L^* and pH in meat balls made by normal and woody breast meat, however, they did not use severe WB fillets and that can explain the different results. Fresh chicken

sausage made of 100% WB had the highest cook loss % compared to the rest of the treatments, which were not different across them ($P \geq 0.05$). In addition, correlation values showed that pH ($r = 0.15$, P – value <0.0001) and lightness ($r = - 0.004$, P – value <0.0001) were not associated with to cook loss. Chen et al. (2017), found similar cook yield in meatballs made with normal and woody breast meat. Tijare et al. (2016) and Bowker et al. (2018), also reported that marinated severe WB breast fillets had higher cook loss compared to normal fillets. In a recent study performed at the same time, Caldas-Cueva and Owens (2019) developed deli-loaves made with normal, moderate and severe WB in 33% increments and found that cook loss increased as the WB increased in the formulation. These results indicate that deli-loaves formulated with 100% severe WB could have a negative impact on the product quality and final yield.

The data from the TPA parameters are summarized in Table 18. Hardness, cohesiveness and springiness are the primary parameters related to forces of attraction between particles of food that oppose disintegration (Munoz et al., 1992). Resilience is how well the product “fights to regain its original height” (Texture Technologies, 2019). The results in the study showed a minor gradual decrease in hardness and cohesion in deli-loaves made with severe WB, however, no significant differences were found among the treatments ($P \geq 0.05$). On the other hand, deli loves made with 100% WB had the lowest springiness and resilience values compared to the control ($P \leq 0.05$). These results were similar to Chen et al. (2017) who also reported lower springiness values in meat balls made with woody breast meat. These results indicate that the inclusion of

more than 75% WB in the formulation of deli-loaves may cause a lack of binding ability and therefore, affect the texture profile of the final product.

3.5. Conclusions

Fresh chicken sausage can be formulated with up to 75% WB without affecting the consumer perception towards the texture of the product. Although consumer sensory analysis should be performed in future studies, the combination of 25% and 75% normal and severe WB fillets could be a suitable option in the formulation of chicken deli-loaves without affecting the texture and yield of the final product. Considering the growing demand for convenient and ready to eat products, the utilization of severe WB could have a potential economic contribution to the poultry industry as compared to its current downgraded status.

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4. EVALUATION OF GROWTH PRODUCTION FACTORS AS PREDICTORS OF THE INCIDENCE AND SEVERITY OF WHITE STRIPING AND WOODY BREAST IN BROILER CHICKENS

4.1. Abstract

White stripping (WS) and woody breast (WB) have been previously associated with older and heavier birds. However, there is limited information supporting the association between these two muscle conditions and growth parameters. The objectives of this study were 1) to investigate the relationship between WS and WB using different growth production factors and 2) to propose a predictive model that uses growth production factors to investigate the incidence and severity of WS and WB. A combined database of 4,332 broilers pooled from seven research experiments conducted from 2016 to 2017 at Texas A&M University was used in this study. Parameters such as sex (females and males), age (4 wk, 6 wk, and 8 wk), specie (standard vs. high breast yield), live weight categories (500g increments) and, breast weight categories (250g increments) were evaluated and included in the model. Results in this study showed that WS was more likely to be present in non-WB fillets than WB present in non-WS breast fillets. The association between WS and WB outcomes suggests a moderate relationship between the ranks of both outcome variables ($\rho = 0.57$, $P < 0.0001$). To understand the relationship and predictive power of the study variables, a proportional odds logistic ordinal regression model was fit to each outcome. Variables such as age, live weight and sex were not as important as breast weight and species in the severity prediction of WS

and WB. Butterfly fillets above 750 g and high breast yielding strains were more likely associated with higher severity of WS and WB myopathy scores. No post-hoc variable selection was performed. Both models show good discrimination. The WS model produced an uncorrected AUC of 0.739, with a bootstrap corrected estimate of 0.736. The WB model produced an uncorrected AUC of 0.753 and a bootstrap corrected estimate of 0.752. Therefore, growth production factors analyzed in this study indicated that there is a moderate relationship between WS and WB and are jointly predictive of the severity of WS and WB. Potentially other factors not included in this study may play a major role in the relationship of these myopathies.

4.2. Introduction

Chicken meat is a popular protein around the world. A recent consumer survey showed that versatility and healthy properties were the main reasons consumers preferred chicken to other protein sources (Neth and Parker, 2018). Since 2012 there has been an increased demand for fresh and deli chicken products and the trends indicate further growth in this area over the next few years (National Chicken Council, 2018; Neth and Parker, 2018).

In order to fulfill the growing demand and make the production more efficient, selection criteria of broiler chickens have been changing through genetic selection, management practices, and nutrition (Bodle et al., 2018; Zuidhof et al., 2014). However, several studies have shown that genetic selection for high breast development in broilers has led to muscle fiber hypertrophy, which is causing structural, functional and metabolic changes (Dransfield and Sosnicki, 1999; Kuttappan et al., 2012b, 2013; Petracci and Cavani, 2012; Velleman and Nestor, 2003).

White striping (**WS**) and woody breast (**WB**) are recent meat quality issues found in fast growing heavy-broilers (Sihvo et al., 2014). The exact etiology of these conditions remains unknown. However, some of the quality issues associated with them are lower water-holding capacity, higher cook loss percentage, lower marinade pick-up percentage and differences in mechanical and sensory texture (higher values of crunchiness, chewiness and fibrousness) compared to normal fillets (Aguirre et al., 2018; Mudalal et al., 2014; Soglia et al., 2017). Recently, the Food Safety and Inspection Service (2018) sent disposition instructions for WS and WB poultry conditions. This

Notice explains that the severe inflammatory tissues associated with WS and WB must be trimmed during inspection. These new dispositions plus the consumer's unwillingness to eat WS/WB meat is causing an economic impact for the poultry industry (Kuttappan et al., 2012a). In this regard, it is imperative to explore new alternatives to identify the cause of the incidence and severity of these conditions.

Predictive models are widely used in clinical studies as an important decision-making tool across various disease procedures in medicine (Aziz et al., 2016; McGirt et al., 2017; Michaelson et al., 2011). For example, McGirt et al. (2017) created predictive models for the efficient selection of patients, based on patient specific factors that need spinal surgery. Predictive models, trained on the appropriate data, can be a key decision-making tool. The following study pooled data sets from several nutritional studies performed at Texas A&M University in order to investigate the relationship between WS and WB using different growth production factors and to propose a predictive model that uses growth production factors to investigate the incidence and severity of WB and WS.

4.3. Materials and Methods

4.3.1. Database

All the projects included in the database used in the present study were approved by the Texas A&M's Institutional Animal Care and Use Committee. A combined database of 4,332 broilers from seven different research experiments conducted during 2016 to 2017 at Texas A&M University were used in this study. Broilers from each study were conventionally processed under similar conditions in a pilot scale processing

facility at Texas A&M University. Upon completion of the debone process, breast fillets were weighed, palpated, and scored for WS and WB. Breast fillets were scored for WS using normal (0) without any white lines, mild (1), moderate (1.5), and severe (≥ 2.5) as described by Owens and Alvarado (2015). The total number of observations for each category was: 0 (782), 0.5 (186), 1 (1725), 1.5 (973), 2 (561), 2.5 (93), 3 (4). Breast fillets presenting WB were categorized according to Tijare et al. (2016) as normal (0) without any hardness, mild (1) hardness present mainly on the cranial region, moderate (2) hardness throughout the fillet with some flexibility in the mid-caudal region, and severe (3) hardness in the cranial/caudal region with no flexibility. The total number of observations in each category was: 0 (973), 0.5 (125), 1 (1591), 1.5 (2), 2 (1193), 3 (440). In addition, breast fillets were scored in 0.5 increments, when necessary, and due to small cell counts some of the cells were combined. One person was in charge of the WS and WB categorization for all the nutritional studies.

The distribution of the birds is presented in Table 19 and Table 20. Factors analyzed for the predictive model included: sex, age, species, live weight categories, and breast weight categories. The full data set consisted of 4,332 observations. A complete case analysis was performed and observations with missing outcome or covariate values were excluded from each analysis. Ultimately, 4,320 observations were used for the WS analysis and 4,321 observations for the WB analysis. Thus, only minimal information was lost due to the exclusion of observations with missing data.

4.3.2. Statistical Analysis

Frequency Analysis and Correlation Coefficient Values. Frequency analysis was used to investigate how overall, and each variable was distributed within each WS and WB severity score. The monotonic association between both myopathies scores was tested using Spearman's rank correlation coefficient. Correlation coefficients between 0.10 to 0.39 were considered as weak, values between 0.40 to 0.69 were moderate, and values between 0.70 to 0.89 were strong (Schober et al., 2018).

Predictive Models. Since outcomes were not equally spaced ordinal ranks (i.e. WB = 2 is not necessarily 2 times as bad as WB = 1, WB = 3 is not necessarily 3 times as bad as WB = 1, etc.), proportional odds ordinal logistic regression models would provide an appropriate and parsimonious modelling method to investigate the relationship between predictors and outcomes. P-values were derived using Wald statistics. No variable selection was performed. Variable importance was measured by how each variable contributed to the overall chi-square. The probabilities of having a WS and WB > 1 for two hypothetical broilers were calculated to provide an example use of the models.

Model Validation. Model Calibration was investigated using calibration plots and a discrimination measure of model performance was computed using a bootstrap resampling method to estimate the likely performance of the model on a new sample of broilers. Discrimination was expressed as a generalized AUC (area under the curve), which measures how well the model can tell the outcome levels apart. A value of 0.5

indicates no predictive discrimination and a value of 1.0 indicates perfect discrimination (Harrell et al., 1996). The analysis was performed in R using the rms package (Harrel Jr., 2018). $P < 0.05$ was considered statistically significant.

4.4. Results

4.4.1. Frequencies and Correlation Coefficients Values

Frequency analysis of the occurrence of each WS scores within each WB scores category is presented in Figure 1. Data suggested that 50.2% of normal WB fillets (WB = 0) exhibited no WS scores. As WB severity scores increased, there was a decreasing trend of fillets that did not present WS (WS = 0). Whereas, there is a slightly increasing trend of severe scores of WS with the increased severity of WB scores. Likewise, it was estimated that 98.4% of severe WB fillets (WB = 3) presented some degree of WS, where 28.4 % were normal to mild, 67.5% moderate, and 10.2% severe. In addition, frequency analysis of the occurrence of each WB scores within each WS scores category is shown in Figure 2. Results suggested that 62% of normal WS fillets (WS = 0) presented no WB. Among that breast fillets containing severe WS scores, 0% were normal, 13.4% mild, 40.2 % moderate and 46.4% had severe WB scores. Similar trends were found compared to the WB frequency analysis.

The association between WS and WB outcomes suggests a moderate monotonic relationship between the ranks of both outcome variables ($\rho = 0.57$, $P = < 0.0001$).

4.4.2. Predictive Models

To better understand the relationship between predictors (variables) and outcomes (WS and WB), a proportional odds ordinal logistic regression model was fit to each outcome to determine the predictive ability of the study predictor variables. Statistical models for WS and WB are presented in Table 21. Variables sex ($P = 0.995$) and live wt category (P - value 0.07) in the WS model, and sex (P - value 0.753), age ($P = 0.707$) and live wt category ($P = 0.08$) in the WB model were not significant. Since the primary objective of the study was to assess whether and to what degree each outcome could be predicted by the predictor variables, there is no need to simplify the model by performing variable selection. The full model consists of only five variables and is not greatly simplified by removing any of them. In addition, automatic variable selection via P - values, although widely used in procedures such as stepwise regression, can be dangerous and lead to incorrect inferences if not done correctly (Heinze et al., 2017 and Whittingham et al., 2006).

The importance of each predictor included in each model is illustrated in Figure 3 for WS and Figure 4 for WB, respectively. Each plot shows how important each variable was measured by how much it contributes to the overall Wald chi-square statistic. The x-axis is the amount of the chi-square statistic the variable accounts for. The higher the amount, the higher is the importance of the predictor in the model. By this measure breast wt was the most important predictor in the severity of WS, and breast wt together with species in WB. Whereas, the least important for both outcomes was sex.

Whether factors increase or decrease the odds of moving into a higher level of the outcomes and by how much are shown in the Table 22. Variables with odds ratio > 1 increase the odds of having a higher myopathy severity, odds ratio = 1 does not affect the odds of the outcome, and variables with odds ratio < 1 decrease the odds (are protective). Importantly, these measures give the increase in the odds ratio for a 1-unit increase in the predictor variable. Results obtained from this study showed that the main growth production factor associated with the increase in severity of WB and WS is breast weight. In particular breast fillets above 750 g were 4.28 times (95% CI, 3.69 to 4.96) in WB and 3.59 (95% CI, 3.09 to 4.16) times in WS more likely to have higher myopathy severity scores compared to lighter fillets ($P = < 0.0001$). Whereas, fillets above 1000 g were associated with higher (highest OR) odds of increased WB (OR, 23.92; 95% CI, 15.91 to 35.97) and WS (OR, 13.90; 95% CI, 9.31 to 20.76) severity ($P = < 0.0001$). On the other hand, fillets weighing less than 500 g were 0.25 (95% CI, 0.16 to 0.37) times in WB and 0.37 (0.25 to 0.56) times in WS less likely to present higher severity myopathy scores ($P = < 0.0001$). Furthermore, in the WS model, 6 wk (OR, 3.08; 95% CI, 1.88 to 5.03; $P = < 0.0001$) and 8 wk (OR, 2.21; 95% CI, 1.23 to 3.94; $P = 0.007$) of age were associated with greater probability of higher severity of WS scores. As for species, results showed that specie B (OR, 1.34; 95% CI, 1.17 to 1.53; $P = < 0.0001$) and C (OR, 1.44; 95% CI, 1.04 to 1.99; $P = 0.02$) which are bred to be high breast yielding broilers, were associated with higher odds of increasing WS severity scores. Live wt categories and sex were not associated with the increase in WS severity scores ($P > 0.05$). As for the WB model, the second most important attribute was specie,

specie B (OR, 2.47; 95% CI, 2.15 to 2.83) and C (OR, 2.49; 95% CI, 1.81 to 3.42) were associated with higher odds of presenting increased severity of WB scores (P – value < 0.0001). In live wt categories, only broilers weighing > 2500 - < 3000 g (OR, 1.64; 95% CI, 1.05 to 2.56) were associated with higher odds of having higher WB severity scores (P = 0.02). Although the rest of the categories showed OR higher than 1, the confidence intervals contain the relative risk of 1.00, therefore the association between WB and age categories was not considered significant which is confirmed by the $P > 0.05$. Age categories and sex also showed no association with the increase in severity scores of WB ($P > 0.05$).

4.4.3. Hypothetical Examples

Based on the proposed model two hypothetical examples were generated and their predicted probabilities of having a score of WS and WB > 1 were computed. As detailed in the Table 23, hypothetical Broiler A is a 4 wk old male with a standard breast yield B, 2200 g live weight, 750 g breast weight. The baseline score for WS and WB was 1, respectively. The average scores of WS and WB are the scores multiplied by the probability of a broiler with the provided characteristics falling into that category added together. As a result, the average WS score was 1.01 and for WB was 1.61. The probability of being in a category above the baseline score in WB was 32.9 % and 21.6 % for WS. These results stand in contrast to those of Broiler B (Table 4), who had an average WS score of 1.73 and 2.37 WB and the probability of having scores above the baseline were higher (WB 89.6 %, WS 84.2 %).

4.4.4. Model Performance and Validation

Both models show good discrimination (Figure 5 and 6). The WS model produced an uncorrected AUC of .739, and a bootstrap corrected estimate of .736. The WB model produced an uncorrected AUC of .753 and a bootstrap corrected estimate of .752. The small difference between uncorrected and corrected estimates suggests that the models are not overfitting the data (Harrell et al., 1996). This is a promising sign for the model's ability to perform well on similar but new samples of data.

The calibration plots suggest good calibration for all levels of each outcome (Figures 5 to 10) except $WS \geq 2.5$ (Figure 11). This plot suggests slight overfitting (the model is providing probabilities that are higher than what would be expected from the observed data). This is due to the relatively small number of observations in the highest levels of the outcome. There are only 97 observations with $WS = 2.5$. Overall, the accuracy of the model's prediction probabilities is good (Harrell et al., 1996). Therefore, the predicted probabilities can likely be trusted in all cases except when trying to predict whether an observation will fall into the highest categories.

4.5. Discussion

4.5.1. Predictors and Outcomes

This study investigated the relationship between WB and WS, and used growth production factors to develop a predictive model for the severity of both muscle conditions. Data demonstrated that WS was approximately 12% more likely to be present in non-WB fillets, than WB present in non-WS breast fillets. These results are in

agreement with Griffin et al. (2017), who investigated the progressive changes and abnormal clinical presentation during P. major post hatch growth and reported that WS was the first myopathy to be present in breast fillets at day 16.

Correlation coefficients demonstrated that there is a positive moderate correlation between WS and WB myopathy scores. In a recent study, Bowker et al. (2019) investigated the association between WS/WB and found similar results ($r_s = 0.55$, $P < 0.0001$) in 2,600 breast fillets collected from a commercial processing plant. The results obtained also demonstrate that regardless of the growing method (commercial or in a controlled research environment) the association between both myopathies is similar. The proportional odds ordinal logistic regression model obtained in the current study demonstrated that breast weight was the most important predictor in the severity of WB and WS. In specific, fillets higher than 750 g have an increased probability of having higher severity myopathy scores and this probability is even higher in breast fillets above 1000 g. On the other hand, 63% of butterfly fillets weighing less than 500 g are less likely to be associated with increased severity WS scores, and 75% with WB scores. These support previous research where moderate and severe degrees of WS were found in heavier fillets (Kuttappan et al., 2013). In addition, Griffin et al. (2017) documented the macroscopic changes occurring with age/growth in the breast muscle and found that breast muscle yield was the primary predictor for the severity of WB. It has been hypothesized that accelerated muscle growth reduced the interstitial space between the epimysium (connective tissue sheath) and the P. major muscle leading to muscle damage (Sihvo et al., 2014; Soglia et al., 2015).

The data showed a strong positive correlation between breast wt and live wt ($\rho = 0.78$; $P = <0.0001$), and live wt and age ($\rho = 0.75$; $P = < 0.0001$). Care should be used when individually interpreting the odds ratios provided by the models. For instance, when breast weight is removed from the model the effect of live weight goes from having lower probability of increasing the severity of the outcome (protective against risk) to increasing risk. Thus, it is only jointly that these estimated odds ratios can be used to predict the outcomes. Additionally, variable importance is conditional on the variables in the model. This means that variables might not look important because another variable is included, but will become so if that variable is excluded. For instance, if breast weight is excluded then age and live weight become relatively more important. Therefore, considering the high correlation between these variables, the results from the present study showed that overall live wt and age can influence the severity of WS and WB myopathy scores. Similar results can be observed in the frequency analysis (Tables 19 and 20), where normal fillets gradually decrease with the increase in age and live wt. Moreover, in a previous study, Kuttappan et al. (2013) and Petracci et al. (2013) reported that heavier birds and thicker breast fillets are most likely to show WS than lighter birds. These results are also consistent with Griffin et al. (2017) which reported that the first documented case of WB was at day 23, and more severe characteristics were observed from day 30 onwards including thick white striations.

Species is the second and third more important variable in the severity of WB and WS respectively. High-breast-yield species had significantly higher odds of having worse severity myopathy scores relative to the standard-breast-yield species.

Furthermore, high-breast-yield species seemed to have more impact on the severity of WB score compared to WS score. Analogous to our results, Bailey et al. (2015) reported that genetic selection has a role in the expression of both myopathies; however, it is not the main driving force. Moreover, Kuttappan et al., (2013) and Trocino et al. (2015) reported that differences between species could be influenced by growth pattern. In addition, results in this study showed that regardless of the specie there is some degree of WS and/or WB present in breast fillets.

The proportional odds ordinal logistic regression model obtained from this study also showed that sex was not significant variable in the prediction of WS and WB (Table 21). Moreover, it also had significantly lower odds of increasing both myopathies severity (Table 22). Even though there is a large disparity in females and males sample size, results from this study are similar to previous research where gender had no significant effect on WS occurrence (Kuttappan et al. 2013).

4.5.2. Application to Practice

The prediction model described provides a probability of having WS and WB in each severity category using growth production factors. The hypothetical case scenarios represent examples of how the combination of different variables predict better or worse outcomes. The application of the proposed model is that companies having data sets could plug in values for variables in this model and get predicted probability in the way that the hypothetical examples provided do. Moreover, the obtained information can be

used as an important decision tool before placing new birds. Therefore, this tool holds the potential to reduce the incidence and severity of WB and WS.

4.5.3. Study Limitations and Strengths

The potential limitations of the current study can be associated to the combined data across experiments. Any inconsistencies in the protocol or deviations could make datasets not suitable for pooling. It is possible that variables not measured and therefore, not included in the model will play a significant role in the prediction of WS/WB. Moreover, missing data could affect the performance of the model in discriminating accurately between observed and predicted outcomes. In this regard, future research should be performed to include more variables in the model in order to improve the performance and accuracy.

On the other hand, the proposed predictive model provides values as starting point to help companies in the decision-making process and optimize outcomes in new flocks of birds. The AUC values for the WB and WS model was 0.797 and 0.766 respectively, which reflects a good discrimination index.

4.6. Conclusions

Using this predictive model, it was demonstrated that there is a positive moderate association between WB and WS. Growth production factors analyzed in this study significantly influence the severity of WS and WB myopathy scores. Most importantly, novel predictive models constructed with data sets hold a potential to reduce the severity of WB and WS by identifying important modifiable factors ante-mortem. Finally,

potentially other factors not included in this study can play a role in the predictive model of WB and WS severity and further research should be conducted.

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5. CONCLUSIONS

The study of the WS and WB muscle conditions have become a challenge for the poultry industry. Three studies were performed from a processing perspective to find alternatives for the utilization of breast meat affected by severe WB, and to understand how growth production factors influence the incidence and severity of WS and WB myopathies.

The combination of RS + STPP in marinade brines provided better results than STPP by itself in improving the cook yield of whole muscle breast fillets affected by severe WB. However, color, pH and the instrumental texture profile of WB remained different than NOR fillets. The combination of CF + STPP in marinade brines showed the highest 24 h post-marinade retention among the ingredients, even though CF was similar to STPP, its 24 h retention values were slightly higher. As for cook loss, WB CF showed the same cook loss performance as NOR CF, and NOR STPP. Likewise, in the TPA severe WB fillets injected with CF + STPP exhibited similar hardness values compared to NOR CF + STPP, and similar springiness and chewiness compared to WB CF and NOR CF + STPP. Overall RS+STPP (hot set), and CF and CF + STPP (cold set) can be used to improve WHC of not only severe WB fillets but NOR fillets as well. Moreover, CF can be used as a clean label ingredient to replace STPP. Additional research needs to be done using a consumer panel to know if consumers can detect differences among the treatments.

Comminuted products reduce the negative effects of severe WB in the texture profile and meat quality parameters. Up to 75% of severe WB can be used in the manufacturing of fresh chicken sausage without negatively affecting color parameters, cook yield and consumer perception. Likewise, results indicated that up to 75% of severe WB can be used in the manufacturing of chicken deli-loaves without affecting color, cook loss and texture. It was also demonstrated that moisture and pH do not change with the comminution process since both variables gradually increased with the increase percentage of WB in sausage and deli-loaves.

White striping and woody breast are positively correlated however their relationship is moderate. It is more likely to find some degree of WS in non-WB filets than some degree of WB in non-WS filets. The proportional odds ordinal regression model used in this study indicated that breast wt was the most important factor influencing the incidence and severity of WS and WB myopathies. Butterfly fillets above 750g were more likely to have higher severity scores than lighter fillets. Butterfly fillets below 500 g were less likely to be associated with increased myopathy scores. Live wt and age were not as important as breast wt in the prediction model, however, breast wt is highly correlated to live wt, and live wt is highly correlated to age, therefore, both factors can influence the severity of WS and WB. High-breast-yield species showed higher probability of having worst severity scores compared to standard breast yield species. Sex had lower odds of increasing both myopathy scores. Overall, the growth production factors included in the prediction model significantly influence the incidence and severity of WS and WB. The application of the proposed model is that companies

having data sets could plug in values for variables in this model and get predicted probability for both myopathy scores. Therefore, the obtained information can be used as an important decision tool before placing new birds. Future research should be performed to include more variables in the model in order to improve the performance and accuracy.

APPENDIX A

TABLES

Table 1. Meat quality characteristics of non-marinated severe WB (mean and pooled SEM)¹

Meat type	Drip loss %	*L	*a	*b	pH
Normal	1.31 ^b	57.79 ^b	1.15	5.32 ^b	5.82 ^b
Woody	1.56 ^a	58.71 ^a	1.2	5.74 ^a	6.04 ^a
Pooled SEM	0.09	0.70	0.08	0.52	0.01

^{a,b}Different letters indicate significant differences among the means in the same column ($P \leq 0.05$)

¹Measurements were taken at 24h postmortem

Table 2. Effect of rice starch (RS) on the pick-up yield (PUY), color and pH of severe WB meat (lsmeans and pooled SEM)

Meat type	PUY	*L	*a	*b	pH
Normal	15.24 ^a	53.30 ^b	0.87 ^b	5.16 ^b	6.07 ^b
Woody	10.04 ^b	55.71 ^a	1.33 ^a	5.75 ^a	6.20 ^a
Pooled SEM	0.22	1.52	0.33	0.47	0.01
Treatment					
STPP	13.79 ^a	53.38 ^b	0.58 ^b	4.97 ^b	6.17 ^a
RS + STPP	11.50 ^b	55.64 ^a	1.62 ^a	5.95 ^a	6.11 ^b
Pooled SEM	0.22	1.52	0.33	0.47	0.01
<i>P</i> – value					
Type	< 0.0001	< 0.0001	0.0005	0.0208	< 0.0001
TRT	< 0.0001	< 0.0001	< 0.0001	0.0001	0.0008
Type x TRT	0.32	0.35	0.43	0.64	0.96

^{a,b}Different letters indicate significant differences among the means in the same column ($P \leq 0.05$)

Table 3. Effect of rice starch (RS) on marinade retention (RET) and cook loss % of severe WB meat (lsmeans \pm SEM)

Type, Treatment	RET 20 min	RET 2 h	RET 24 h	Cook loss %
Normal, RS + STPP	99.27 \pm 0.13 ^a	99.04 \pm 0.16 ^a	99.16 \pm 0.22 ^a	13.04 \pm 1.90 ^c
Woody, RS + STPP	98.17 \pm 0.25 ^b	97.77 \pm 0.20 ^b	97.34 \pm 0.25 ^b	14.64 \pm 1.97 ^c
Normal, STPP	97.82 \pm 0.25 ^{bc}	97.52 \pm 0.20 ^{bc}	97.38 \pm 0.25 ^b	24.83 \pm 2.02 ^b
Woody, STPP	97.51 \pm 0.15 ^c	97.13 \pm 0.17 ^c	96.50 \pm 0.22 ^c	33.40 \pm 1.91 ^a
	<i>P</i> – value			
Type	0.6795	0.0003	< 0.0001	< 0.0001
TRT	0.0004	< 0.0001	< 0.0001	< 0.0001
Type x TRT	0.0017	0.0025	0.0059	< 0.0001

^{a,b}Different letters indicate significant differences among the means in the same column ($P \leq 0.05$)

Table 4. Effect of citrus fiber (CF) on the PUY, 24 h retention (RET) and color (a*, b*) of severe WB meat (lsmeans and pooled SEM)

Meat type	PUY	RET 24 h	*a	*b
Normal	17.01 ^a	97.88 ^a	0.45 ^b	5.62
Woody	11.41 ^b	96.39 ^b	1.03 ^a	5.84
Pooled SEM	0.26	0.31	0.25	0.47
Treatment				
CF + STPP	13.58 ^b	97.74 ^a	0.91	5.91 ^a
CF	15.26 ^a	96.96 ^b	0.72	6.32 ^a
STPP	13.79 ^b	96.71 ^b	0.58	4.97 ^b
Pooled SEM	0.32	0.31	0.26	0.48
<i>P</i> – value				
Type	< 0.0001	< 0.0001	< 0.0001	0.2395
TRT	0.0010	< 0.0001	0.0585	< 0.0001
Type x TRT	0.0996	0.0511	0.7013	0.1882

^{a,b}Different letters indicate significant differences among the means in the same column ($P \leq 0.05$)

Table 5. Effect of citrus fiber (CF) on the 20 min and 2h retention (RET), lightness, pH, and cook loss % of severe WB meat (Ismeans \pm SEM)

Type, treatment	RET 20 min	RET 2 h	*L	pH	Cook loss %
Normal, CF+ STPP	98.80 \pm 0.24 ^a	98.85 \pm 0.31 ^a	53.78 \pm 1.50 ^{ab}	6.14 \pm 0.04 ^{bc}	15.01 \pm 2.01 ^d
Normal, CF	98.54 \pm 0.23 ^a	97.74 \pm 0.32 ^b	53.64 \pm 1.50 ^b	5.89 \pm 0.04 ^d	24.22 \pm 2.01 ^c
Normal, STPP	97.45 \pm 0.23 ^b	97.39 \pm 0.32 ^{bc}	52.04 \pm 1.50 ^c	6.10 \pm 0.04 ^c	23.74 \pm 2.00 ^c
Woody, CF + STPP	97.89 \pm 0.23 ^b	97.35 \pm 0.32 ^{bc}	54.31 \pm 1.50 ^{ab}	6.20 \pm 0.04 ^{ab}	29.32 \pm 2.00 ^{ab}
Woody, CF	97.88 \pm 0.23 ^b	97.07 \pm 0.31 ^c	54.90 \pm 1.50 ^a	6.07 \pm 0.03 ^c	26.56 \pm 1.98 ^{bc}
Woody, STPP	97.46 \pm 0.23 ^b	96.92 \pm 0.32 ^c	54.73 \pm 1.51 ^{ab}	6.24 \pm 0.03 ^a	32.80 \pm 2.00 ^a
	<i>P</i> – value				
Type	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001
TRT	< 0.0001	< 0.0001	0.0114	< 0.0001	< 0.0001
Type x TRT	0.0003	0.0005	0.0017	0.0082	< 0.0001

^{a,b}Different letters indicate significant differences among the means in the same column (*P* - value \leq 0.05)

Table 6. Effect of rice starch (RS) on the Texture Profile Analysis of severe WB meat (Ismeans and pooled SEM)

Meat type	Hardness	Springiness	Cohesiveness	Chewiness
Normal	3.24 ^b	0.79 ^b	0.81 ^b	2.06 ^b
Woody	4.24 ^a	0.81 ^a	0.82 ^a	2.77 ^a
Pooled SEM	0.10	0.11	0.09	0.05
Treatment				
STPP	3.77	0.80	0.81 ^b	2.41
RS+ STPP	3.71	0.81	0.82 ^a	2.42
Pooled SEM	0.10	0.11	0.09	0.05
	<i>P</i> - value			
Type	< 0.0001	0.0137	0.0227	< 0.0001
TRT	0.6369	0.5027	0.0281	0.9685
Type x TRT	0.6674	0.2868	0.6148	0.4361

^{a,b}Different letters indicate significant differences among the means in the same column (*P* - value \leq 0.05)

Table 7. Effect of citrus fiber (CF) on the Texture Profile Analysis of severe WB meat (Ismeans and pooled SEM)

Meat type	Cohesiveness		
Normal	0.80 ^b		
Woody	0.82 ^a		
Pooled SEM	0.09		
Treatment	Cohesiveness		
CF + STPP	0.83 ^a		
STPP	0.81 ^b		
CF	0.79 ^c		
Pooled SEM	0.09		
<i>P</i> – value			
Type	0.0006		
TRT	< 0.0001		
Type x TRT	0.6344		
Treatment, Type	Hardness	Springiness	Chewiness
Woody, CF	4.47 ^a	0.81 ^a	2.78 ^{ab}
Woody, STPP	4.31 ^a	0.81 ^a	2.81 ^a
Woody, CF + STPP	4.21 ^{ab}	0.81 ^a	2.78 ^{ab}
Normal, CF + STPP	3.78 ^b	0.80 ^a	2.48 ^b
Normal, STPP	3.24 ^c	0.79 ^a	2.01 ^c
Normal, CF	3.13 ^c	0.77 ^b	1.85 ^c
Pooled SEM	0.22	0.09	0.12
<i>P</i> – value			
Type	< 0.0001	0.0006	< 0.0001
TRT	0.1707	0.0086	0.0005
Type x TRT	0.0013	0.0004	0.0003

^{a,b}Different letters indicate significant differences among the means in the same column (*P* - value ≤ 0.05)

Table 8. Effect of severe woody breast on quality attributes of raw ground meat (mean and Pooled SEM, n = 12)

Meat type	*L	*a	*b	pH
Woody breast	62.44 ^a	1.13	9.26 ^a	6.22 ^a
Normal	58.57 ^b	0.84	8.12 ^b	5.99 ^b
Pooled SEM	0.64	0.32	0.31	0.02

^{abc}Different letters indicate significant differences among the means in the same column (P - value ≤ 0.05)

Table 9. Effect of the inclusion of different percentages of severe woody breast (WB) on the quality attributes of raw chicken sausage (mean and pooled SEM, n = 30)

WB (%)	*L	*a	*b	pH
0%WB	64.67	7.06	21.94	6.20 ^a
25% WB	61.39	7.37	21.49	6.27 ^{ab}
50% WB	65.89	6.49	21.18	6.31 ^b
75% WB	63.11	6.50	19.21	6.32 ^c
100% WB	65.96	6.85	18.93	6.35 ^d
Pooled SEM	1.28	0.55	1.16	0.008

^{abc}Different letters indicate significant differences among the means in the same column ($P \leq 0.05$)

Table 10. Effect of the inclusion of different percentages of severe woody breast (WB) on the quality attributes of cooked chicken sausage (mean and pooled SEM, n = 30)

WB (%)	*L	*a	*b	pH
0%WB	67.10	4.93	19.86	6.37 ^b
25% WB	66.13	5.80	20.22	6.39 ^b
50% WB	66.90	5.29	19.71	6.43 ^{ab}
75% WB	69.09	4.40	17.31	6.44 ^{ab}
100% WB	64.87	5.11	18.04	6.48 ^a
Pooled SEM	1.14	0.32	1.02	0.02

^{abc}Different letters indicate significant differences among the means in the same column ($P \leq 0.05$)

Table 11. Effect of the inclusion of different percentages of severe woody breast (WB) on moisture, fat, and cook loss percentage of chicken sausage (lsmeans and pooled SEM)

WB (%)	Moisture (%)	Fat (%)	Cook loss (%)
0% WB	52.46 ^c	28.77 ^a	11.41 ^a
25% WB	55.63 ^{ab}	25.80 ^b	8.08 ^{cd}
50% WB	54.33 ^b	26.61 ^{ab}	9.41 ^{bc}
75% WB	55.36 ^{ab}	26.89 ^{ab}	7.60 ^d
100% WB	56.05 ^a	25.77 ^b	10.13 ^{ab}
Pooled SEM	0.46	0.64	0.42

^{abc}Different letters indicate significant differences among the means in the same column ($P \leq 0.05$)

Table 12. Effect of the inclusion of different percentages of severe woody breast (WB) on the texture profile analysis (TPA) of chicken sausage (Ismeans and pooled SEM, n = 45)

WB (%)	[¥] Hardness	[¤] Cohesion	[§] Springiness	[¶] Chewiness
0% WB	5.52 ^a	0.88	501.08	24205.38 ^a
25% WB	5.13 ^{ab}	0.90	511.93	23616.93 ^{ab}
50% WB	4.75 ^{ab}	0.89	488.69	20551.56 ^{ab}
75% WB	4.59 ^b	0.90	491.33	20211.36 ^b
100% WB	5.07 ^{ab}	0.89	500.82	22260.23 ^{ab}
Pooled SEM	0.22	0.005	12.32	1159.44

^{abc}Different letters indicate significant differences among the means in the same column ($P \leq 0.05$)

[¥]Hardness = Maximum force during the first compression

[¤]Cohesiveness = Area of work during the second compression divided by the area of work during the first compression

[§]Springiness = Distance of the detected height during the second compression divided by the original compression distance

[¶]Chewiness = Gumminess * Springiness

Table 13. Demographic frequencies for consumers (n = 80) in sensory analysis of chicken sausage

Question	Number of Respondents	Percentage of Respondents
<i>Gender</i>		
Female	41	51.25
Male	39	48.75
<i>Age</i>		
20 years or younger	8	10.00
21-25	35	43.75
26-35	20	25.00
36-45	4	5.00
46-55	6	7.50
56-65	6	7.50
66 years and older	1	1.25
<i>Ethnicity</i>		
Caucasian	58	72.50
Latino	13	16.25
Asian	7	8.75
African-American	1	1.25
Native American	1	1.25

Table 14. Effect of the inclusion of different percentages of severe woody breast (WB) on the sensory consumer attributes¹ of chicken sausage (Mean and pooled SEM)

WB (%)	Overall	Color	Flavor	Texture	Juiciness
0% WB	6.55	5.88	6.41	6.23 ^a	6.61
25% WB	6.21	5.64	6.20	5.88 ^{ab}	6.67
50% WB	6.01	5.90	6.03	5.81 ^{ab}	6.75
75% WB	6.15	5.86	6.16	5.90 ^{ab}	6.86
100% WB	5.85	5.77	6.06	5.20 ^b	6.53
Pooled SEM	0.22	0.21	0.23	0.24	0.18

^{abc}Different letters indicate significant differences among the means in the same column (P - value ≤ 0.05)

¹Attributes measured where 0 = dislike extremely and 9 = like extremely

Table 15. Categories of terms, example of comments, and the frequencies of occurrence for deli-loaves formulated with 0, 25, 50, 75 and 100% WB

Category	Open-ended comments	Treatment					<i>P</i> – value
		0% WB	25% WB	50% WB	75% WB	100% WB	
Appearance (%)							
Like appearance	Good color, less greasy	19	11	17	19	33	0.010
Dislike appearance	Very greasy, little fat, pale color	18	26	19	18	19	0.681
Texture (%)							
Like texture	Good mouthfeel, crispy, good consistency, easy to bite	25	21	24	16	14	0.319
Dislike texture	Inconsistent, spongy, chewy, gelatinous, tough, too tender	15	16	16	20	33	0.635
Moisture (%)							
Like moisture	Juicy	17	21	19	22	21	0.938
Dislike moisture	Dry, very juicy	20	26	20	26	9	0.049
Flavor (%)							
Like flavor	Flavorful	19	24	19	18	21	0.890
Dislike flavor	poor flavor, funky taste, unpleasant aftertaste	13	19	24	19	24	0.382

Table 16. Effect of the inclusion of different percentages of severe woody breast (WB) on the quality attributes of raw chicken deli-loaves (lsmeans and pooled SEM, n = 75)

WB (%)	*L	*a	*b	pH
0%WB	59.16 ^b	1.48 ^a	4.96 ^b	6.10 ^c
25% WB	59.03 ^b	1.04 ^{ab}	5.38 ^b	6.13 ^{bc}
50% WB	60.18 ^{ab}	0.78 ^{bc}	5.66 ^{ab}	6.15 ^b
75% WB	61.26 ^a	0.56 ^{cd}	5.77 ^{ab}	6.19 ^a
100% WB	60.69 ^a	0.24 ^d	6.49 ^a	6.20 ^a
Pooled SEM	0.25	0.06	0.18	0.006

^{abc}Different letters indicate significant differences among the means in the same column ($P \leq 0.05$)

Table 17. Effect of the inclusion of different percentages of severe woody breast (WB) on the quality attributes of cooked chicken deli-loaves (lsmeans \pm SEM)

WB (%)	*L	*a	*b	pH	Cook loss %
0% WB	71.11 \pm 0.51 ^c	2.22 \pm 0.20 ^{bc}	9.69 \pm 0.25 ^{bc}	6.27 \pm 0.03 ^d	17.48 \pm 0.30 ^b
25% WB	72.44 \pm 0.49 ^{bc}	3.05 \pm 0.20 ^a	9.38 \pm 0.25 ^c	6.29 \pm 0.03 ^c	17.71 \pm 0.28 ^b
50% WB	73.45 \pm 0.49 ^{ab}	2.46 \pm 0.20 ^{ab}	9.75 \pm 0.25 ^{bc}	6.31 \pm 0.03 ^{bc}	17.34 \pm 0.28 ^b
75% WB	73.68 \pm 0.49 ^{ab}	1.76 \pm 0.20 ^{cd}	10.49 \pm 0.25 ^{ab}	6.34 \pm 0.03 ^b	17.37 \pm 0.30 ^b
100% WB	75.22 \pm 0.49 ^a	1.55 \pm 0.20 ^d	10.83 \pm 0.25 ^a	6.36 \pm 0.03 ^a	19.06 \pm 0.28 ^a

^{abc}Different letters indicate significant differences among the means in the same column ($P \leq 0.05$)

Table 18. Effect of the inclusion of different percentages of severe woody breast (WB) on the texture profile analysis (TPA) of chicken deli-loaves (Ismeans and pooled SEM)

WB (%)	[¥] Hardness	[*] Resilience	^² Cohesiveness	[§] Springiness	[¶] Chewiness
0% WB	25.43 ± 1.43	0.78 ± 0.01 ^a	84.86 ± 1.40 ^a	19587.18 ± 1270.26 ^a	40381.95 ± 8452.58
25% WB	25.24 ± 1.43	0.77 ± 0.01 ^{ab}	82.93 ± 1.40 ^{ab}	19382.55 ± 1270.26 ^{ab}	55820.62 ± 8452.58
50% WB	24.42 ± 1.43	0.75 ± 0.01 ^{bc}	78.86 ± 1.35 ^b	18345.46 ± 1270.26 ^{ab}	45695.34 ± 8452.58
75% WB	22.99 ± 1.43	0.75 ± 0.01 ^{bc}	79.97 ± 1.45 ^{ab}	17111.56 ± 1270.26 ^{ab}	44966.13 ± 8452.58
100% WB	22.97 ± 1.43	0.74 ± 0.01 ^c	80.22 ± 1.52 ^{ab}	16721.85 ± 1270.26 ^b	44043.08 ± 8452.58

^{abc}Different letters indicate significant differences among the means in the same column (*P* - value ≤ 0.05)

[¥]Hardness = Maximum force during the first compression

^{*}Resilience = Upstroke energy of the first compression divided by the downstroke energy of the first compression

^²Cohesiveness = Area of work during the second compression divided by the area of work during the first compression

[§]Springiness = Distance of the detected height during the second compression divided by the original compression distance

[¶]Chewiness = Gumminess * Springiness

Table 19. White striping frequency analysis with respect to the growth production variables included in the analysis

Variable	WS Frequency					Total
	0	1	1.5	2	2.5	
Sex (%)						
Female	237 (50)	186 (40)	30 (6)	14 (3)	3 (1)	470
Male	545 (14)	1725 (45)	943 (24)	547 (14)	90 (2)	3853
Age (%)						
4 wk	342 (61)	218 (39)	0	0	0	560
6 wk	410 (12)	1637 (46)	929 (26)	472 (13)	93 (3)	3541
8 wk	30 (14)	56 (26)	44 (20)	89 (41)	0	219
Species (%)						
Standard breast yield A	559 (29)	860 (44)	311 (16)	163 (8)	44 (3)	1937
High breast yield B	31 (16)	104 (55)	33 (17)	18 (9)	4 (2)	190
High breast yield C	192 (9)	947 (43)	629 (29)	380 (17)	49 (2)	2197
Live weight (%)						
< 2000 g	274 (70)	116 (30)	2 (1)	0	0	392
> 2000- < 2500 g	99 (37)	153 (57)	15 (6)	2 (1)	0	269
> 2500- < 3000 g	122 (23)	272 (50)	94 (17)	46 (9)	5 (1)	539
> 3000- < 3500 g	200 (13)	732 (47)	386 (25)	205 (13)	43 (3)	1566
> 3500- < 4000 g	80 (6)	589 (43)	424 (31)	226 (17)	44 (3)	1363
> 4000 g	6 (3)	49 (25)	52 (27)	82 (42)	5 (3)	194
Breast weight (%)						
< 500 g	287 (67)	132 (31)	6 (1)	2 (0)	0	427
> 500 - < 750 g	394 (21)	1036 (55)	322 (17)	99 (5)	17 (1)	1868
> 750 - < 1000 g	96 (5)	722 (39)	602 (32)	386 (21)	68 (4)	1874
> 1000 g	3 (2)	19 (13)	33 (23)	74 (52)	12 (9)	141

Table 20. Woody breast frequency analysis with respect to the growth production variables included in the analysis

Variable	WB Frequency				Total
	0	1	2	3	
Sex (%)					
Female	258 (55)	158 (33)	44 (9)	10 (2)	470
Male	715 (19)	1558 (41)	1151 (30)	430 (11)	3854
Age (%)					
4 wk	338 (60)	197 (35)	22 (4)	3 (1)	560
6 wk	619 (17)	1464 (41)	1090 (31)	372 (11)	3545
8 wk	16 (7)	55 (25)	83 (38)	65 (30)	219
Species (%)					
Standard breast yield A	734 (38)	771 (40)	327 (17)	103 (5)	1937
High breast yield B	43 (23)	77 (41)	59 (31)	11 (6)	190
High breast yield C	196 (9)	868 (40)	809 (37)	324 (15)	2197
Live weight (%)					
< 2000 g	265 (68)	119 (30)	7 (2)	1 (0)	392
> 2000- < 2500 g	124 (46)	117 (43)	26 (10)	3 (1)	270
> 2500-< 3000 g	149 (28)	191 (35)	146 (27)	53 (10)	539
> 3000-< 3500 g	282 (18)	681 (43)	449 (29)	154 (10)	1566
> 3500-< 4000 g	148 (11)	559 (41)	488 (36)	168 (12)	1363
> 4000 g	5 (3)	49 (25)	79 (41)	61 (31)	194
Breast weight (%)					
< 500 g	302 (71)	118 (28)	8 (2)	0	428
> 500 - < 750 g	555 (30)	861 (46)	371 (20)	91 (5)	1878
> 750 - < 1000 g	115 (6)	727 (39)	755 (40)	277 (15)	1874
> 1000 g	0	9 (6)	60 (43)	72 (51)	141

Table 21. Statistical model for white striping and woody breast outcomes

White Striping Model, Wald Statistics			
Variables	Chi-Square	D.F.	<i>P</i> – value
Sex	0.00	1	0.995
Age category	24.78	2	< .0001
Species	20.49	2	< .0001
Live weight category	10.18	5	0.070
Breast weight category	366.89	3	< .0001
TOTAL	1306.76	13	<.0001

Woody Breast Model, Wald Statistics			
Variables	Chi-Square	D.F.	<i>P</i> – value
Sex	0.10	1	0.753
Age category	0.69	2	0.707
Species	178.06	2	< .0001
Live weight category	9.83	5	0.080
Breast weight category	496.68	3	< .0001
TOTAL	1347.95	13	<.0001

Table 22. White striping and woody breast odds ratio (OR) for variables included in the model

Dependent variable:	White Striping		
	OR	95 % CI	<i>P</i> - value
Sex = m	1.0	0.77 to 1.29	0.99
Age = 6 wk	3.08	1.88 to 5.03	<0.0001
Age = 8 wk	2.21	1.23 to 3.94	0.007
Specie B	1.34	1.17 to 1.53	<0.0001
Specie C	1.44	1.04 to 1.99	0.02
Live wt = < 2000 g	0.85	0.53 to 1.35	0.5
Live wt = > 2500 - < 3000 g	0.98	0.63 to 1.53	0.93
Live wt = > 3000 - <3500 g	1.25	0.79 to 1.96	0.32
Live wt = > 3500 - <4000 g	1.34	0.84 to 2.13	0.2
Live wt = > 4000 g	1.68	0.96 to 2.92	0.06
Breast wt = < 500 g	0.37	0.25 to 0.56	<0.0001
Breast wt = > 750 - < 1000 g	3.59	3.09 to 4.16	<0.0001
Breast wt = > 1000 g	13.90	9.31 to 20.76	<0.0001
Dependent variable:	Woody Breast		
	OR	95 % CI	<i>P</i> - value
Sex = m	1.04	0.80 to 1.35	0.75
Age = 6 wk	1.07	0.65 to 1.76	0.77
Age = 8 wk	0.95	0.53 to 1.70	0.88
Specie B	2.47	2.15 to 2.83	<0.0001
Specie C	2.49	1.81 to 3.42	<0.0001
Live wt = < 2000 g	1.33	0.83 to 2.13	0.22
Live wt = > 2500 - < 3000 g	1.64	1.05 to 2.56	0.02
Live wt = > 3000 - <3500 g	1.38	0.87 to 2.16	0.16
Live wt = > 3500 - <4000 g	1.29	0.81 to 2.05	0.26
Live wt = > 4000 g	1.34	0.76 to 2.32	0.3
Breast wt = < 500 g	0.25	0.16 to 0.37	<0.0001
Breast wt = > 750 - < 1000 g	4.28	3.69 to 4.96	<0.0001
Breast wt = > 1000 g	23.92	15.91 to 35.97	<0.0001

Table 23. Predicted probability of WB and WS>1 derived from a predictive model for 2 hypothetical broilers (1 and 2)

Variables	Broiler 1	Broiler 2
Sex	Male	Male
Age	4 wk.	8 wk.
Species	Standard breast yield	High breast yield
Live Weight	2200	4000
Breast Weight	750	1000
Baseline WS/WB	Score 1	Score 1
Average WS Level	1.01	1.73
Average WB Level	1.61	2.365
Probability of WS > 1	21.6%	84.2%
Probability of WB > 1	32.9%	89.6%

APPENDIX B

FIGURES

Figure 1. Frequency of breast fillets expressed as percentage of woody breast scores within each white striping scores category

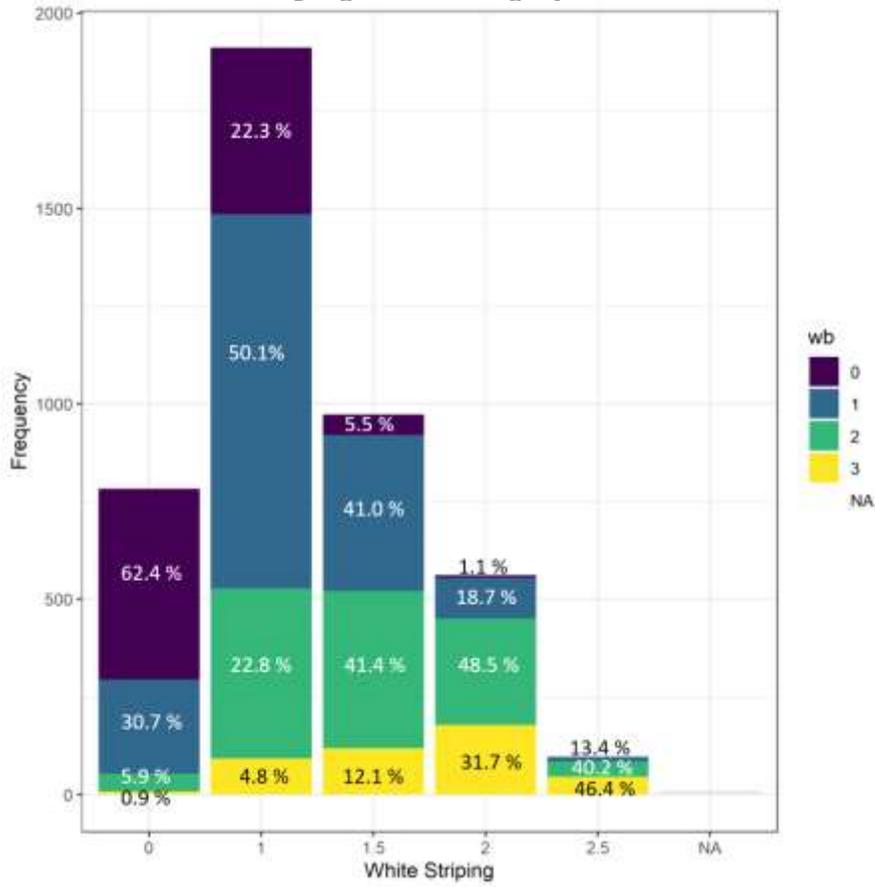


Figure 2. Frequency of breast fillets expressed as percentage of white striping scores within each woody breast scores category

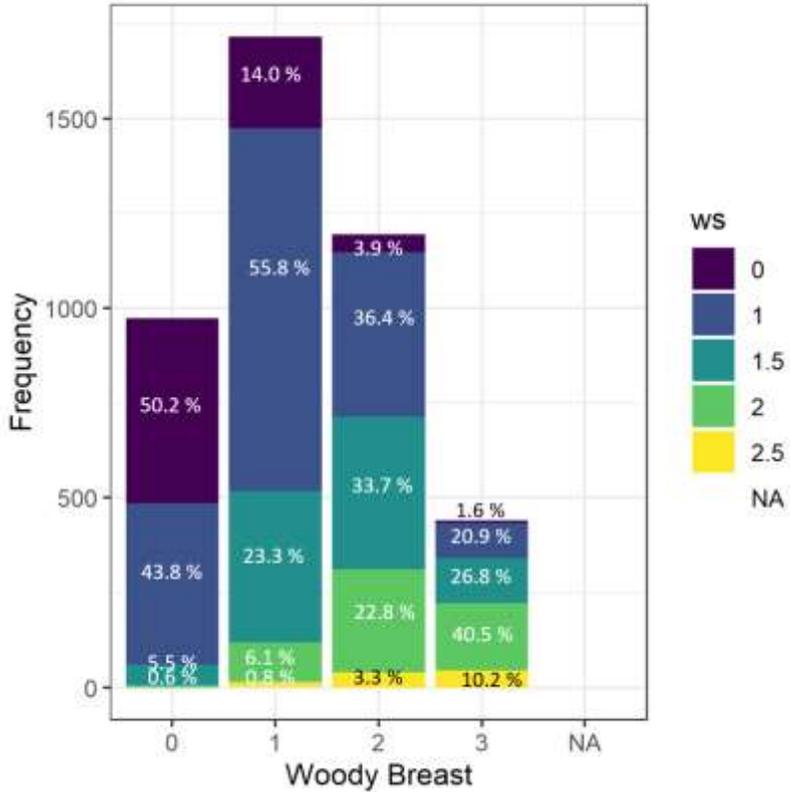


Figure 3. White striping model variable importance

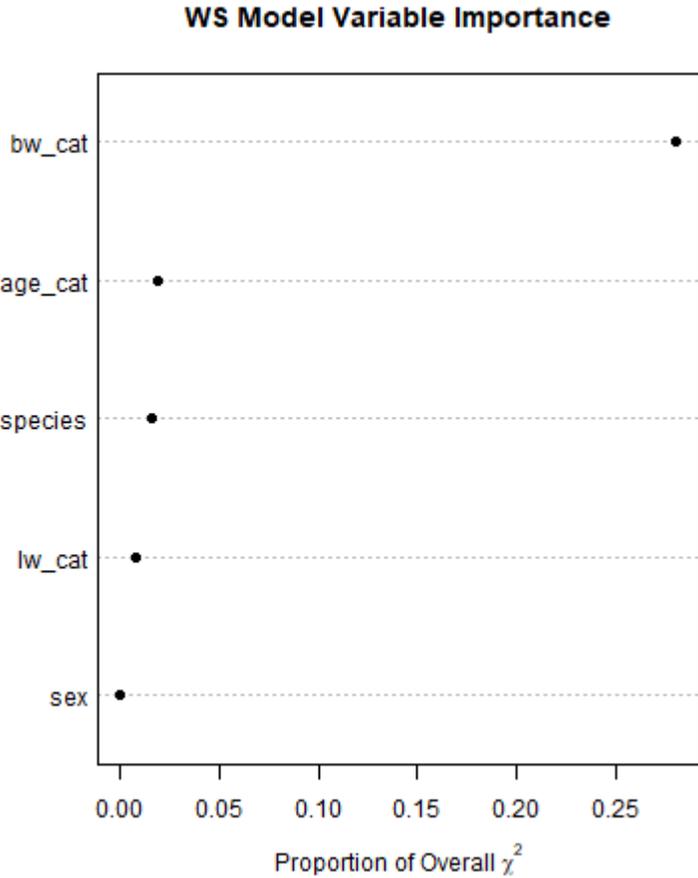


Figure 4. Woody breast model variable importance

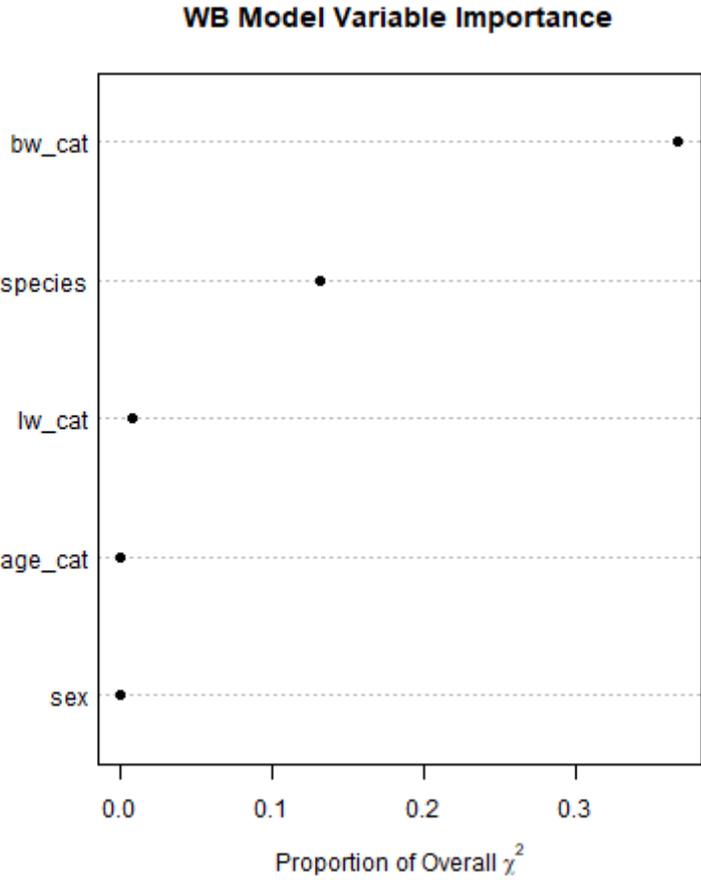


Figure 5. Model validation for predicted probability of woody breast ≥ 1

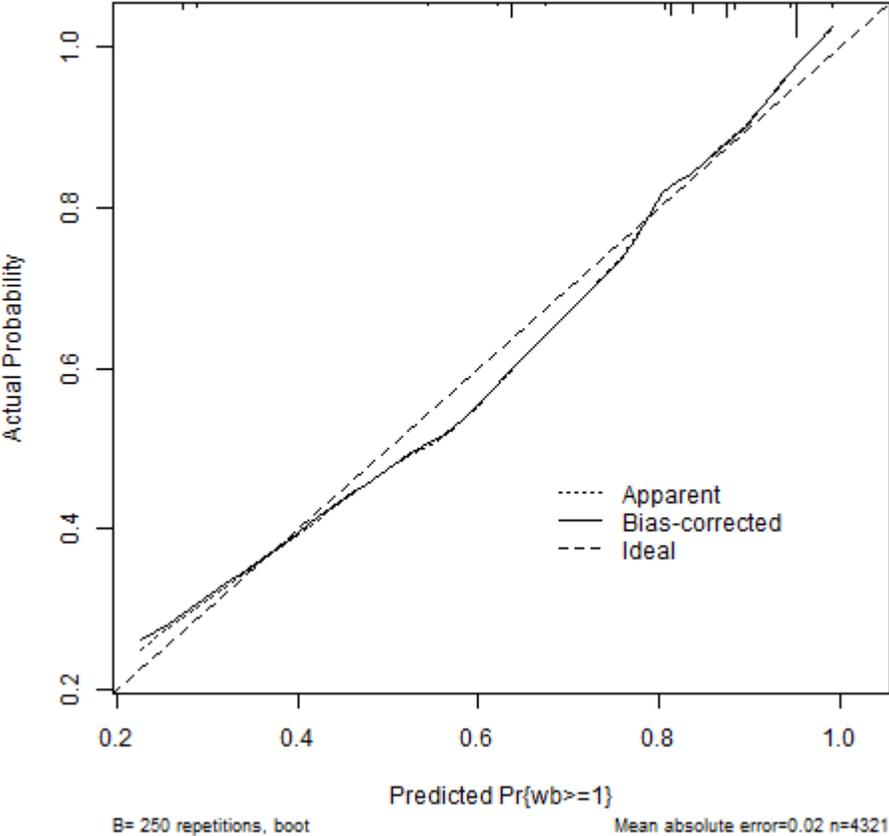


Figure 6. Model validation for predicted probability of woody breast ≥ 2

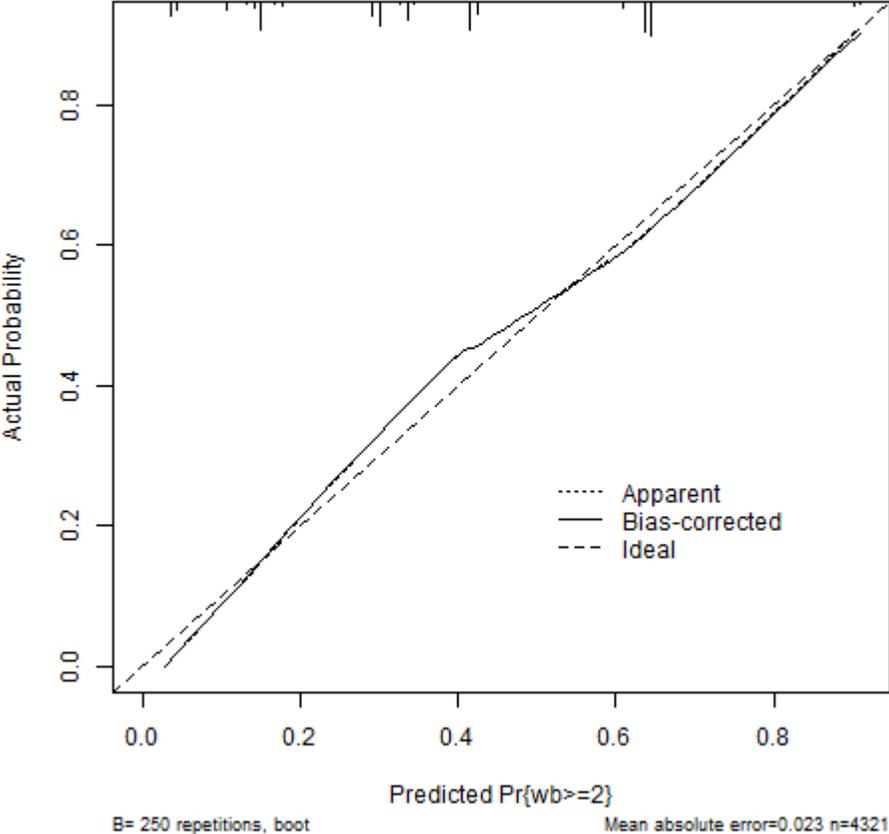


Figure 7. Model validation for predicted probability of woody breast ≥ 3

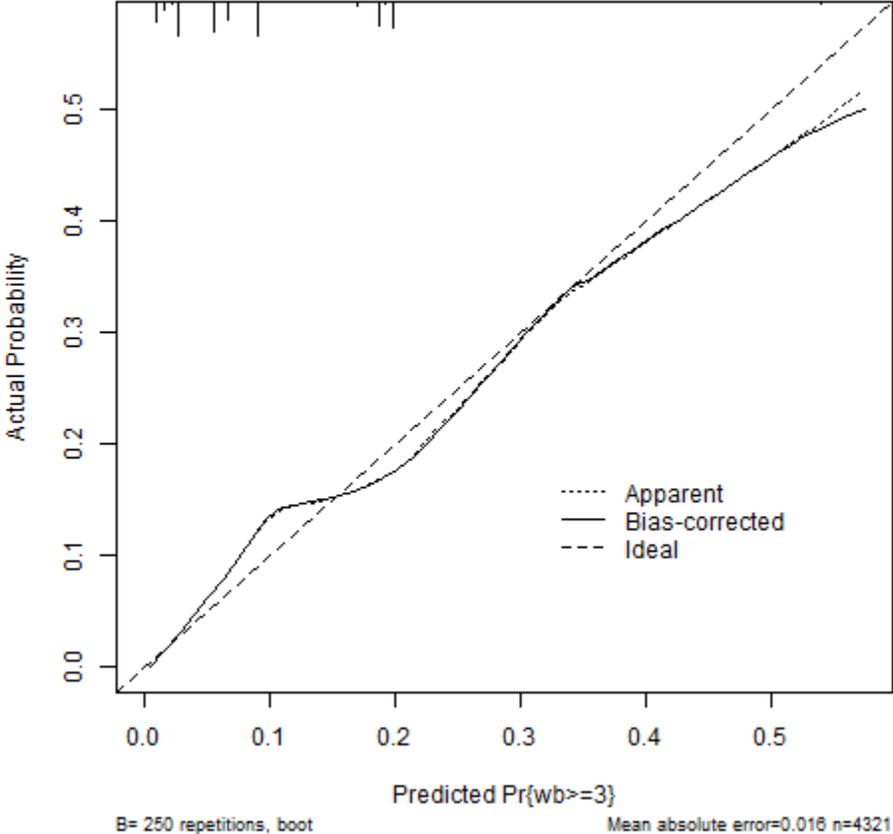


Figure 8. Model validation for predicted probability of white striping ≥ 1

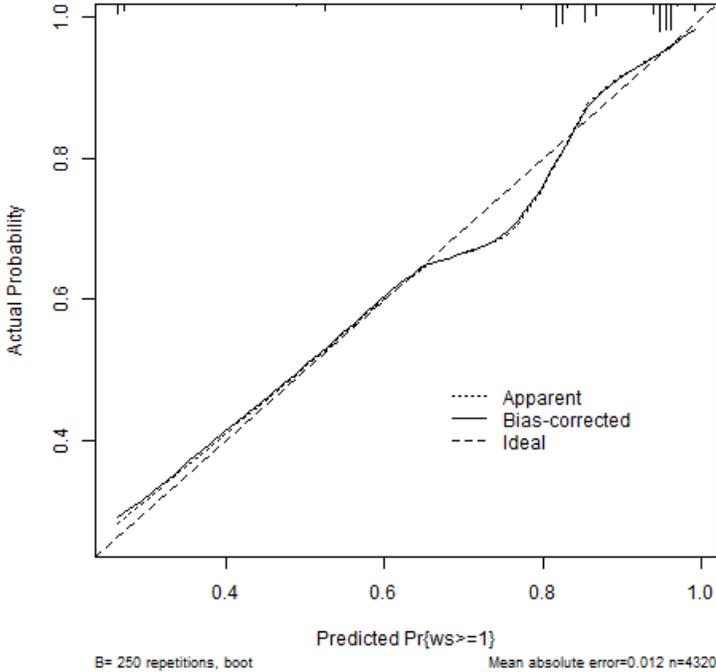


Figure 9. Model validation for predicted probability of white striping ≥ 1.5

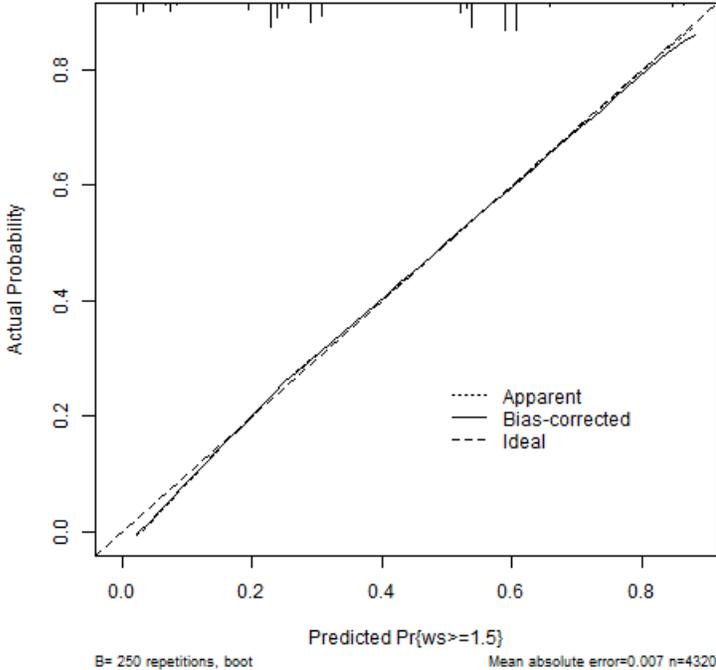


Figure 10. Model validation for predicted probability of white striping ≥ 2

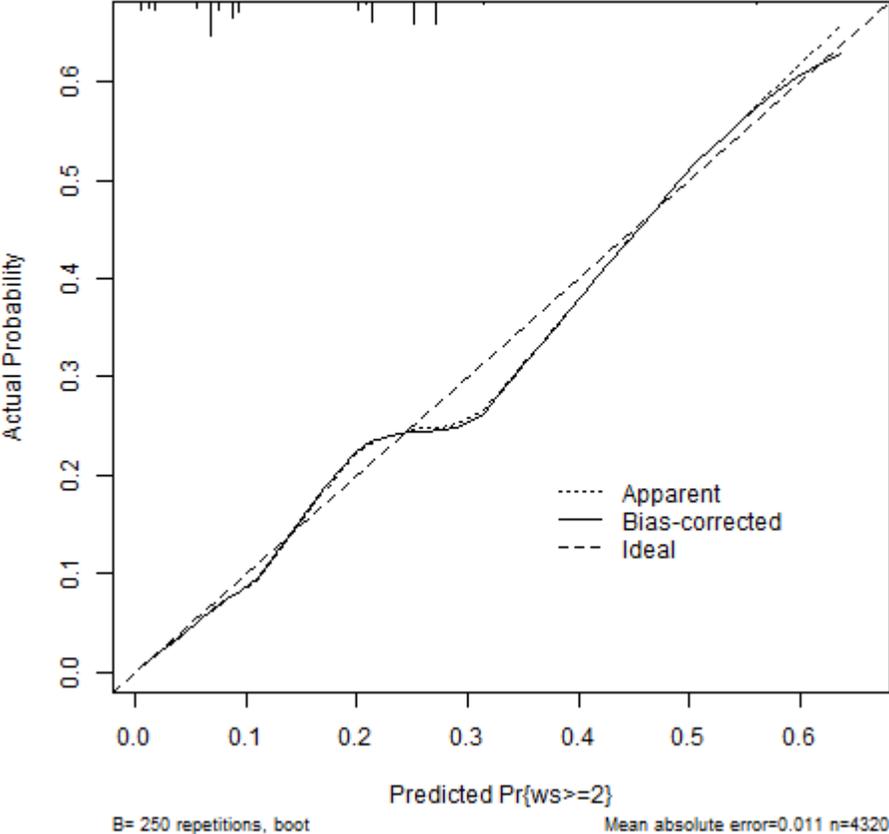
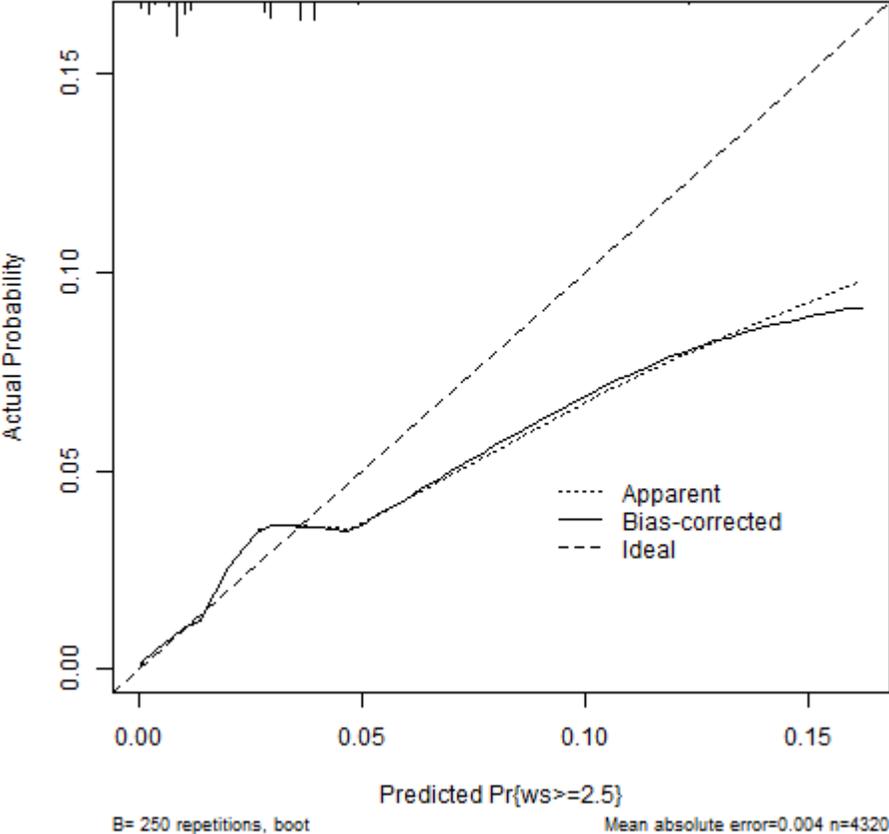


Figure 11. Model validation for predicted probability of white striping ≥ 2.5



APPENDIX C

FORMS

Code _____

INSTRUCTIONS

Thank you for your participation in this study. Your assistance is very much appreciated. The objective of this study is to evaluate chicken breast products. Please take your time and evaluate the samples given to you carefully. Please proceed at your own rate.

This sampling will take you about 30 minutes and you will be eating **A total of 2-8 samples**. Please answer the following questions as completely as possible. If you have any questions, please ask the monitor for assistance.

1. Samples will be served one at a time.
2. Between samples please cleanse your pallet with a bite of cracker followed by a sip of water.

BOLD LETTERS throughout the questionnaire will give you directions on how to complete the evaluation.

Thank you very much for your help and opinions.



IRB NUMBER: IRB2016-0783M
IRB APPROVAL DATE: 04/28/2017
IRB EXPIRATION DATE: 12/01/2021

Code _____

DEMOGRAPHIC INFORMATION

Please circle each appropriate response.

1. Please indicate your gender.

Male

Female

2. Which of the following best describes your age?

20 years or younger

21 - 25 years

26 - 35 years

36 - 45 years

46 - 55 years

56 - 65 years

66 years and older

3. Please specify your ethnicity.

African-American

Latino or Hispanic

Asian/Pacific Islanders

Native American

Caucasian (non-Hispanic)

Other

4. What cooking method do you prefer to use when cooking chicken? Circle any that apply.

Pan-frying or using a skillet on the stove

Stir Fry

Grilling Outside

Oven Broiling

Oven Baking

Microwave

Electric Appliance (George Foreman Grill or other electric grill)



IRB NUMBER: IRB2016-0783M
IRB APPROVAL DATE: 04/28/2017
IRB EXPIRATION DATE: 12/01/2021

Code _____

INSTRUCTIONS

Please take a bite of cracker followed by a sip of water prior to evaluating the product. Place a mark in the box that represents your answer for each of the following questions.

1. How much do you like or dislike this product **OVERALL**?

<input type="checkbox"/>								
Dislike				Neither				Like
Extremely				Like or Dislike				Extremely

2. How much do you like or dislike of the **COLOR** of this product?

<input type="checkbox"/>								
Dislike				Neither				Like
Extremely				Like or Dislike				Extremely

3. How much do you like or dislike of the **FLAVOR** of this product?

<input type="checkbox"/>								
Dislike				Neither				Like
Extremely				Like or Dislike				Extremely

4. How much do you like or dislike of the **TEXTURE** of this product?

<input type="checkbox"/>								
Dislike				Neither				Like
Extremely				Like or Dislike				Extremely

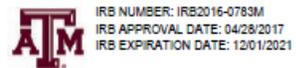
5. How much do you like or dislike the **JUICINESS** of this product?

<input type="checkbox"/>								
Dislike				Neither				Like
Extremely				Like or Dislike				Extremely

5. What do you like about the sample?

6. What do you dislike about the sample?

THANK YOU VERY MUCH FOR YOUR HELP AND OPINIONS.



Texas A&M University Human Research Protection Program
Consent form

Project Title: Chicken meat products

You are invited to take part in a research study being conducted by Dr. Christine Alvarado, a researcher from Texas A&M University. The information in this form is provided to help you decide whether or not to take part. If you decide you do not want to participate, there will be no penalty to you.

Why is this study being done?

To evaluate the sensory quality and consumer acceptance of chicken meat products.

Why am I being asked to be in this study?

You are being asked to be in this study because you eat chicken meat products.

How many people will be asked to be in this study?

Approximately 100 participants will be invited to participate in this study.

What will I be asked to do in this study?

You will be asked to sample a variety of chicken meat products and complete a questionnaire related to each sample. Your participation in this study will last approximately 30 minutes.

Are there any risks to me?

The only risk or discomforts would be from tasting various samples of chicken meat products.

Will there be any cost to me?

Aside from your time, there are no costs for taking part in the study.

Will I be paid to be in this study?

No monetary compensation will be provided.

Will information from this study be kept private?

Efforts will be made to limit the use and disclosure of your personal information, including research study and other records, to people who have a need to review this information. We cannot promise complete privacy. Organizations that may inspect and copy your information include the TAMU HRPP and other representatives of this institution. No identifiers linking you to this study will be included in any sort of report that might be published.

Who may I contact for more information?

You may contact the Principal Investigator, Dr. Christine Alvarado, to tell her about a concern or complaint about this research at 806-438-2293 or calvarado@poultry.tamu.edu.

For questions about your rights as a research participant, to provide input regarding research, or if you have questions, complaints, or concerns about the research, you may call the Texas A&M

Texas A&M University Human Research Protection Program

Consent form

University Human Subjects Protection Program office by phone at 1-979-458-4067, toll free at 1-855-795-8636, or by email at irb@tamu.edu.

What if I change my mind about participating?

This research is voluntary and you have the choice whether or not to be in this research study. You may decide to not begin or to stop participating at any time.

I have read and understand the explanation provided to me. I have all my questions answered to my satisfaction, and I voluntarily agree to participate in this study.

Signature of participant

Date

Dr. Christine Alvarado

Date

