IMPROVEMENT OF CHILLING EFFICIENCY AND PRODUCT QUALITY OF BROILER CARCASSES USING SUB-ZERO SALINE SOLUTIONS FOR CHILLING

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ABSTRACT

Improvement of chilling efficiency and product quality of broiler carcasses using sub-zero saline solutions for chilling

Morgan Michelle Metheny

Sub-zero saline solutions were evaluated for the improvement of chilling efficiency and product quality of broiler carcasses. In this study, four experiments were conducted to chill broiler carcasses using different saline solutions and chilling temperatures in the Meat Processing Center at California Polytechnic State University (Cal Poly, San Luis Obispo, CA) or in the processing plant at Foster Farms (Livingston, CA). In Experiment I, three salt concentrations and solution temperatures (0% NaCl/0.5ºC, 4% NaCl/-2.41ºC, and 8% NaCl/-5.08ºC) were used to chill carcasses. The fillets in brine chilling at sub-zero temperatures showed lower shear forces than the fillets in 0% NaCl control solution. In Experiment II, three salt concentrations (0% NaCl/0.5ºC, 4% NaCl/-2.41ºC, and 8% NaCl/-5.08ºC) were used to chill carcasses with/without pre-chilling in 0% NaCl/0.5ºC or 0% NaCl/14°C. Fillets from the carcasses in 4% NaCl/-2.41ºC significantly improved tenderness ($P < 0.05$), with no significant difference observed for the shear force of 8% NaCl/-5.08ºC, regardless of pre-chilling. In Experiment III, four salt concentrations (0% NaCl/0.5ºC, 1% NaCl/-0.6°C, 2% NaCl/-1.2°C, and 3% NaCl/-1.8°C) were used to chill carcasses. The shear force of fillets decreased as the salt content increased and chilling temperature decreased from 0%NaCl/0.5°C to 3%NaCl/-1.8°C, with the lowest shear force observed in 3% NaCl brine at -1.8°C ($P < 0.05$). The chilling time (90 min) of 3% NaCl was reduced by 25 min (or 22%) compared to water control (115 min), with an intermediate reduction (13 - 17%)

seen for other NaCl solutions (95 – 100 min). Breast fillets showed no significant difference in chilling yield, pH, R-value, and sarcomere length for raw meats as well as in cooking yield and salt content for cooked fillets across all treatments ($P > 0.05$). In Experiment IV, three salt concentrations (0% NaCl/0.5°C, 3% NaCl/-1.8°C, and 4% NaCl/-2.41°C) were used to chill carcasses. The chilling time (55 min) of 4% NaCl was reduced by 35 min (or 39%) compared to the time (90 min) of water control, with an intermediate reduction (11%) seen for 3% NaCl solution. Control fillets in 0% NaCl showed a higher shear force than the fillets in sub-zero brine chilling ($P < 0.05$). Based on these results, broiler carcasses chilled in 4% NaCl/-2.41°C appears to be ideal to improve both chilling efficiency and meat tenderness compared to the carcasses chilled in 0% NaCl/0.5°C.

Keywords: Broiler processing, Brine chilling at sub-zero temperature, Processing efficiency, Product quality

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INTRODUCTION

1.1 Statement of Problem

The poultry industry relies heavily on the use of water in order to produce safe and wholesome products. Environmental and regulatory factors like the California drought, natural and fiscal resources, and the Clean Water Act make it an important priority to improve the efficiency of production, as well as reduce the amount of water needed throughout processing (Cohen & Sonosky, 1962, California Department of Food and Agriculture., 2015).

1.2 Purpose of Study

The purpose of this study was to investigate the effects of various sub-zero brine chilling solutions on broiler carcasses for improving chilling efficiency and meat quality. By reducing the water temperature and required time in the carcass chiller, processors are able to increase processing efficiency and production productivity that can reduce water consumption and processing costs, amongst other factors. For this study, four experiments were conducted using different saline solutions and chilling temperatures compared to conventional ice slurry to evaluate the carcass chilling efficiency, raw meat quality, meat tenderness, and overall processing efficiency of broilers in the Meat Processing Center at California Polytechnic State University (Cal Poly, San Luis Obispo, CA) or in the processing plant at Foster Farms (Livingston, CA).

Chapter 2
LITERATURE REVIEW

2.1 Chilling of Broiler Carcasses

In commercial poultry processing, chilling broilers post-evisceration is required in order to reduce carcass temperature and pathogenic growth of bacteria. Poultry carcass temperatures need to be reduced from 38°C (100°F) to 4°C (40°F) or below within 4 – 8 hours after evisceration, depending on the carcass weight (USDA, 2014). The three most common poultry chilling techniques are water immersion chilling (WIC), air chilling (AC), and evaporative air chilling (or a combination of WIC and AC). Of the different possible chilling processes, WIC is more commonly used in the United States, while European and South American countries typically use AC. It has been agreed that immersion chilling is the most efficient technique in reducing carcass temperature over air chilling and evaporative air chilling (Thompson *et al.*, 1987, Huezo *et al.*, 2007). Immersion chilling is the process of fully immersing carcasses into chilling water at approximately 1-4°C (33.8-39.2°F), contained in standing vats or through in-line processing, in order to quickly reduce the temperature of the bird. Due to water's heat transfer rate being 25 times better than air, immersion chilling allows the carcass temperature to decrease in a shorter time period compared to air chilling (Sams and McKee, 2011). Over time, the incorporation of large in-line chilling tanks led to a rise in production capacity, product yield, operating costs, and food safety (Morris & Associates, 2012).

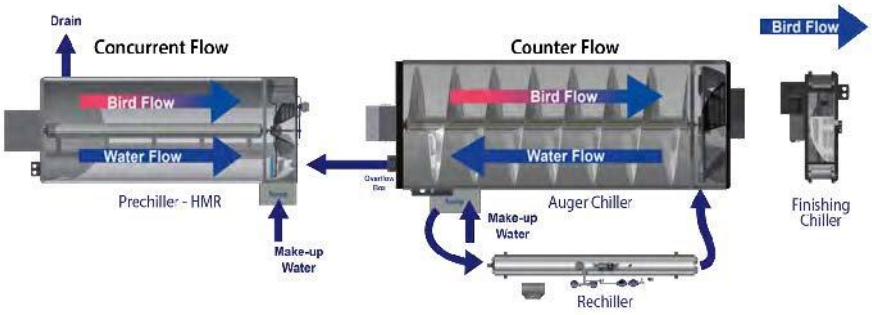

Figure 2.1 Flow of water and broilers during immersion chilling (Source: Barbut).

As seen in Figure 2.1, one or two different chilling tanks can be used to chill birds. In the case of two chilling tanks, the birds at about 38°C (100°F) initially enter a pre-chiller that is set to 7 to 15°C (45 to 59° F) for a cycle of 10 to 15 minutes to allow water absorption, along with added carcass washing. At the end of the pre-chiller, the carcass temperature has dropped to 30-35°C (86-95°F) before entering the main chiller. In the main chiller, water flows in counter current of the carcasses to control the temperature and cleanliness of the water. Chiller tank water ranges from 1°C at the end to 4°C at the start of the tank. Also, the chiller incorporates air flow from the bottom of the tank for consistent water agitation and even distribution of temperature as seen in Figure 2.2. In the case of one chilling tank, the birds are directly placed into the chiller without having the step of pre-chilling.

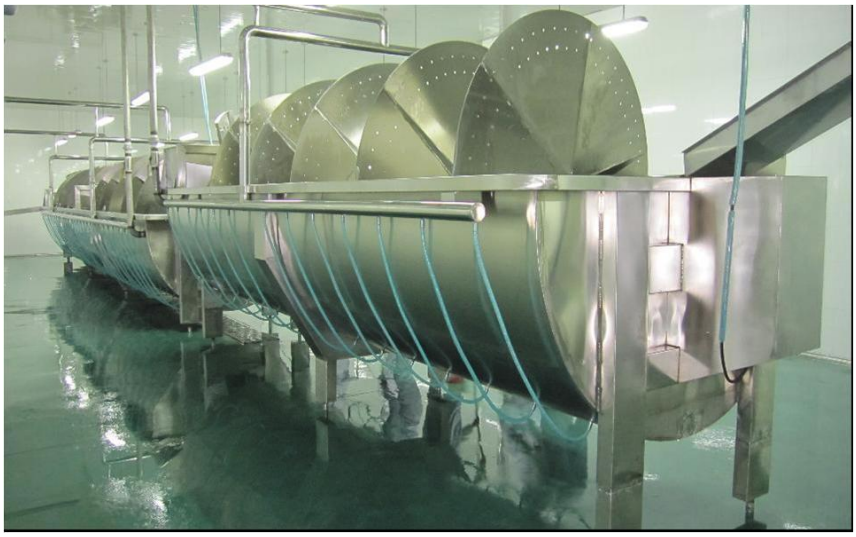

Figure 2.2 Commercial broiler chilling system (Source: Hospibuz).

When the broilers reach the end of the chiller, the carcass temperature will have decreased from 30-38°C (86-100°F) to 4°C (40°F) or below within an hour (Alvarado & Sams, 2002). Although USDA-FSIS regulation allows 4-8 hours for broilers to fully chill to 4°C (40°F) or below, it is typical for the carcasses to reach the proper temperature within 1 to 2 hours post mortem (Owens *et al.*, 2010, USDA, 2014).

2.2 Carcass Chilling Time

In most commercial facilities, the broiler chilling time is much shorter, (typically 1-2 hours) than the allowance of USDA (4-8 hours) (Owens *et al.*, 2010, USDA, 2014). The two most important reasons for quick chilling are to reduce microbial growth and improve processing efficiency (Jeong *et al.*, 2011). Of the various broiler carcass chilling methods, water immersion chilling proves to be the most efficient at reducing the carcass temperature. In a study conducted by Jeong et al. (2011), the internal temperature of carcasses was reduced to 4°C within 60 minutes in WIC, whereas AC took over 120 minutes to chill the carcass. By reducing chilling time, processors are able to increase the

volume of broiler carcasses and potentially maximize the efficiency of broiler processing. In addition, chilling the carcass to 4°C in a shorter amount of time may improve microbial safety of the product and water usage of the plant.

2.3 Saline Solution Chilling

The addition of sodium chloride, or salt, to water depresses the freezing point of the solution in comparison to pure water (Potter *et al.*, 1978). The freezing temperature depression is negatively correlated to the concentration of salt that is within the brine solution. As a result, the higher concentration of salt added the lower the water temperature can reach before freezing. Introducing brine chilling to broiler processing has the potential to improve chilling efficiency by reducing the chilling water temperature below the current commercial standard of 1-4°C (Owens *et al.*, 2010). Another benefit of sub-zero brine chilling is its ability to improve the quality of the chicken muscle. Broiler carcasses that were immersed in 5% NaCl ice slush solutions at -1°C for 4 hours showed an increase in muscle tenderness (Hoey *et al.*, 1983). The increased tenderness is due to the increased water holding capacity and decreased muscle shrinkage (Janky *et al*., 1978). Unfortunately, the addition of NaCl to the chilling water may increase the amount of salt within the broiler carcasses (Hoey *et al.*, 1983).

2.4 Ice Slurry Chilling

The food industry heavily relies on the cooling ability of refrigeration systems. Unfortunately, these cooling systems can result in expensive energy consumption and a negative environmental impact (Ure, 1999). In an effort to reduce the economic and environmental impact of refrigeration systems, the concept of applying ice slurry systems to food processing continues to be explored. An ice slurry can be defined as a number of

small ice crystals within an aqueous solution, typically in the presence of a depressant, such as sodium chloride (Egolf & Kauffeld, 2005). The addition of a depressant allows the solution to reach a freezing temperature below 0°C. A lower freezing point increases the rate of heat transfer between the chilling solution and the product it is intended to chill (Rowe, 2016). The rate of heat transfer in a saline ice slurry can be accelerated up to 5-6 times higher than that of chilled water when it comes to cooling down a warm solid object (Davies, 2004). In addition to an increased rate of heat transfer, ice slurries can also be beneficial when it comes to chilling broilers because the rapid chilling can minimize chemical and enzymatic degradation. Even further, the small ice particles within the slurry may create a scrubbing-like effect on the broiler's skin, thus reducing the bacterial population on the surface (Piñeiro *et al*., 2004). Piñeiro et al. (2014) also indicated that ice slurries as an aqueous suspension of small ice particles are still able to flow through mechanical systems such as pumps, pipes, and tanks. In regards to the poultry industry implementing immersion chilling, it is an added benefit that the ice slurry has the potential to move through equipment smoothly.

2.5 Industrial Impact of Immersion Chilling

According to the USDA's Poultry Production and Value Summary (2016), the United States produced 8.69 billion broilers bringing in $28.7 billion in 2015, which is 2 percent up in number of broilers, yet 12 percent down in revenue from 2014. In regards to live weight, broilers accounted for 53.4 billion pounds in 2015, up 4 percent from 2014. The state of California slaughtered 244 million broilers in 2015 which is about 3 percent of the U.S. total (Baertlein & Hoffstutter, 2015). These high production numbers require an even higher amount of water to fully process the broilers. On average, poultry

processors will use approximately 3.5 to 10.0 gallons of water per bird (North Carolina, 2010). In addition, processing plants are required to allow an overflow rate of 0.25 gal./bird for the scalder and 0.50 gal./bird for the chiller. Overall, processing uses about 76 percent of water during slaughter shifts, followed by 13 percent used during sanitation, and the remaining 12 percent used in downtime (North Carolina, 2010).

The poultry industry's water dependency and impact on their surroundings influenced the Clean Water Act of 1972, which required processors to treat their wastewater to meet environmental water standards in order to keep from negatively affecting nearby ecosystems. Once this Act was introduced, water and wastewater costs increased from $0.33/1000 gal to an average cost of $5.00/1000 gal. This translates to each additional gallon added into production raises the processed bird cost by 0.5 cent (Owens *et al.*, 2010).

These high level water requirements are a considerable concern for poultry processors, especially for those located in California. For example, a large California commercial poultry processor located in Livingston uses 4 to 5 million gallons of water each day (Baertlein & Hoffstutter, 2015). Combined with the drought that has been affecting California the past few years, the amount of water required to run a poultry facility is strenuous on the surrounding communities and environment. With these implications, water use and conservation continues to be a major focus for broiler processors.

2.6 Carcass Chill Yield

Carcass chill yield is the measure of the retained water, comparing the carcass weights before and after the immersion chilling process. Previous studies indicated that carcasses increase in weight by retaining water through the immersion chilling process and that the amount of water retention is directly correlated to chilling time (Bigbee & Dawson, 1963). The majority of the water is trapped between the skin and muscle of the birds, but will lose the gained water during fabrication steps. Carcasses gain up to 11.7% moisture during immersion chilling, but have reported to lose around 5.7% moisture during cut-up processing and an additional 2.1% in storage (Huezo *et al.*, 2007).

Due to the broiler's ability to retain water throughout processing, USDA (2001) published a regulation requiring processors to maintain control and record the amount of water retained within its poultry products. Under this regulation, establishments are required to document the amount of water that is retained as an "unavoidable result of processes used to meet food safety requirements".

2.7 Product Quality

Textural and sensory properties are two important factors that are frequently used to define the quality of the final product. In order to measure the quality of meat, a variety of techniques have been used including instrumental analyses, descriptive sensory analysis, consumer sensory evaluations, or a combination of those (Lyon *et al.,* 1985, Cavitt *et al.,* 2004). Different instrumental analysis procedures may include shear force testing, measure of sarcomere length, pH, or R-value. The breast muscle is typically used for textural analysis due to its high value and popularity, postmortem bio-chemistry, and subsequent fiber characteristics (Lyon *et al.,* 1985).

2.7.1 Shear Force

There is a variety of instrumental techniques used to texturally analyze meat tenderness including Warner–Bratzler Shear Blade, Allo-Kramer "shear compression system (multiple blade)," Texture Profile Analysis (TPA), and Cavitt's razor blade shear method (Cavitt *et al.,* 2004). Since tenderness is a major quality determinant and the most important sensory characteristic for consumers, these techniques are used to numerically identify and evaluate the tenderness of the muscle (Cavitt *et al.,* 2004). Such shearing measurements allow the tenderness of muscles to be analyzed objectively. Shear force is measured by placing the cooked sample's fibers perpendicular to the blade(s) and measuring, in weight or force units, the amount of force required for the blade to shear the sample. The peak amount of force required to penetrate the sample is a measure of the muscle's tenderness (Lyon *et al.,* 1984 & 1986).

The Warner-Bratzler shear method was the first to be developed using a single blade to shear a uniform core of meat and registering the peak load required to shear the meat (Smith *et al.,* 1988). The Allo-Kramer method was developed in the 1950's and is similar to the Warner-Bratzler method, however uses multiple blades to compress then shear the meat instead of a singular blade (Smith *et al.,* 1988). The Texture Profile Analysis (TPA) method evaluates tenderness through a combination of a subjective and objective test. The subjective test uses a trained sensory panel, while the objective test uses a double compression of the sample to mimic chewing compared to the two previous methods (Smith *et al.,* 1988). Lastly, Cavitt's razor blade shear method uses a single razor blade to record the peak force required to shear the surface of the meat sample (Cavitt *et al.,* 2004). Cavitt's method differs from the Warner-Bratzler method because it does not require the sample to be initially cored out before shearing.

2.7.2 pH

After an animal dies, the animal tissue shifts from live muscle tissue to meat, "animal tissues which are suitable for use as food" (Aberle *et al.*, 2012). During this process, the muscle cells switch their metabolic system from the aerobic to anaerobic pathway due to the absence of oxygen available throughout the body (Figure 2.3). In comparison to aerobic metabolism, the anaerobic pathway produces an excess of hydrogen ions and lactic acid when generating energy from glucose. Glycolysis in an anaerobic environment causes two ATP molecules to release 2 hydrogen ions during the process into the cell. Due to an increased concentration of lactic acid and hydrogen ions within cells, the pH of the tissue declines over time after harvest (Aberle *et al*., 2012). After comparing pH values of muscle after harvest, Lyon *et al.* (1984) reported that the pH value declines until it plateaus around 5.9 at 4 hours post-mortem. The four hour sample group also resulted in the most tender meat. It has been concluded that as the pH of the muscle decreases post-mortem, the tenderness of the meat increases (Stewart *et al*., 1984). The inclusion of salt during the chilling process has shown to improve overall tenderness, however several potential issues have come across including the need for special equipment, waste water treatment, and maintaining a low-sodium food (Thompson *et al.*, 1987).

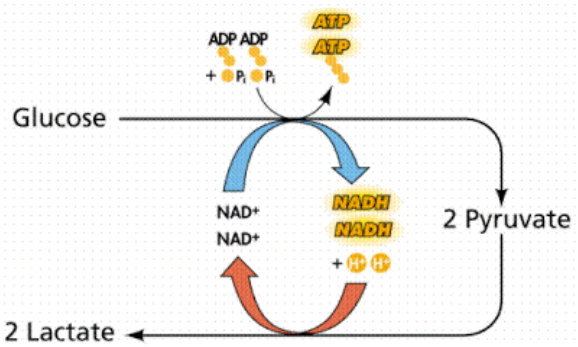

Figure 2.3 Simplified diagram of anaerobic respiration (Source: The A Level Biologist).

2.7.3 R-value

R-value is an indirect measurement of the ATP status in muscle that is calculated as the ratio of inosine and hyposanthine to adenosine (Thompson *et al.,* 1987). R-value is the ratio of the dephosphorylation and deamination of adenosine triphosphate nucleotides (ATP) to the formation of inosinic acid (IMP) (Kahn & Frey, 1971). As observed by Kahn and Frey (1971), the R-value ranges from 0.8 to 1.24, where 0.8 represents pure IMP and 1.24 represents pure ATP. Absorbance ratio was measured by dividing the ATP absorbance value by the IMP absorbance. Upon further review, the samples having an R-value of ~0.8 have produced a greater amount of IMP from ATP due to the onset of rigor mortis. In comparison to the study by Kahn and Frey (1971), our study inverted the ATP/IMP ratio of Kahn and Frey to measure the absorbance ratio, indicating that the higher the R-value the more the ATP breakdown (or less ATP).

2.7.4 Sarcomere Length

Sarcomere length is correlated to the amount of muscle shortening and ultimate tenderness of the meat. A sarcomere is a singular contractile unit within a myofibril. Each sarcomere is composed of an assortment of thick and thin filaments that span from z-line to z-line. The z-line borders of each sarcomere anchor the thin filaments made up of primarily actin proteins. Stacked alongside the thin filaments are thick filaments made up

of myosin proteins that are anchored within the m-line that is located in the center of each sarcomere. During contraction, the myosin filaments contain myosin heads that attach to the actin filaments pulling the actin filaments towards the m-line causing the length of the sarcomere to shorten. Once contraction is complete, the myosin heads release from the actin filaments allowing the sarcomere length to increase during muscle relaxation (Aberle, *et al.,* 2012). Since the overall state of muscle contraction is dependent on sarcomere length, it has been demonstrated that there is a positive relationship between tenderness and sarcomere length (Cross, *et al.,* 1980).

There is a variety of techniques that can be used in order to measure sarcomere length. Such techniques can be grouped together to be either an oil-immersion based method or a laser based method. Oil-immersion methods measure the sarcomere length using phase-contrast microscopy of unstained tissue and recording the measurement through a calibrated eye-piece micrometer. In comparison, the laser method measures the length of the sarcomere by measuring the diffraction pattern that appears when the light penetrates the muscle fibers (Cross, *et al.,* 1980).

Overall, four experiments were conducted in this study to evaluate chilling efficiency, tenderness, and meat quality after chilling broilers in sub-zero brine solutions. By improving the chilling efficiency, the amount of water usage could potentially decrease in addition to processing time, space, and related labor. In regards to tenderness, the shear force results were evaluated with the comparison of pH, R-value, and sarcomere length results due to their correlation with one another.

Chapter 3

EXPERIMENTS I, II, III, & IV: CARCASS CHILLING IN CONTROL AND COLD BRINE SOLUTIONS

3.1 Introduction

The poultry industry relies on the use of water in order to produce safe and wholesome products. Environmental and regulatory factors like natural and fiscal resources and the Clean Water Act make it a priority to improve the efficiency of production, as well as reduce the amount of water required for processing (Cohen & Sonosky, 1962, California Department of Food and Agriculture., 2015). The USDA-FSIS allows commercial facilities to chill broilers to an internal temperature of ≤ 4°C (40°F) within 4 – 8 hours during processing depending on carcass weight, however most facilities chill broilers within 1 -2 hours (Owens et al., 2010, USDA, 2014). Brine chilling has the potential to improve chilling efficiency by reducing the chilling water temperature below the current commercial standard of 1-4°C (Owens *et al.*, 2010).

The quality of boiler carcasses is influenced by chilling time and temperature (Sansawat *et al.*, 2014). Broiler carcasses chilled in 5% NaCl solution at -1°C showed lower shear force values, higher moisture content, higher cooking yield, and more desirable sensory attributes than broilers chilled in an ice slurry at 1°C (Janky *et al.*, 1978). In 1983, Hoey et al. compared breast fillet quality after chilling broilers for 4 hours, with agitation, in 5% NaCl at -1°C or in water at 1°C. The breast fillets in 5% NaCl brine solution at -1°C showed more water and sodium absorption than fillets chilled in water at 1°C. By reducing the chilling temperature of broilers, the breast fillets have shown a reduced amount of rigor shortening, lower textural variability, and improved tenderness (Dunn *et al.*, 1995). Through the reduction of rigor shortening, the myofibrils

have not shortened in a state of contraction resulting in decreased shear force of breast fillets (increased tenderness) (Cross, *et al.,* 1980). In addition to a lower shear force, with a lesser state of contraction the myofibrils have not processed as much ATP to reduce the R-value and pH within the muscle due to anaerobic glycolysis (Kahn & Frey, 1971, Lyon *et al.,* 1984). Furthermore, a lower pH does not promote a higher water holding capacity which reduces the potential tenderness of the breast fillet (Aberle *et al*., 2012).

For this study, four experiments were conducted to evaluate the effects of various salt concentrations and chilling temperatures on chilling efficiency and product quality of broiler carcasses. The purpose of Experiment I was to initially evaluate the effect of sub-zero saline chilling on chilling efficacy and product tenderness with a broad range of brine concentrations and temperatures from 4% NaCl/-2.41°C and 8% NaCl/-5.08°C. In Experiment II, the 0, 4 and 8% solutions were further evaluated, in addition to comparing single chiller and dual chiller scenarios. Following the results of Experiment II, Experiment III was conducted to evaluate a more restricted range of salinity within brine chilling solutions of 0% NaCl/0.5°C, 1% NaCl/-0.6°C, 2% NaCl/-1.2°C, and 3% NaCl/-1.8°C. Lastly, Experiment IV was conducted to directly evaluate the efficacy of 0% NaCl/0.5°C, 3% NaCl/-1.8°C, and 4% NaCl/-2.41°C brine chilling solutions.

3.2 Materials and Methods

3.2.1 Brine Chilling Solution and Brine Ice Preparation

Prior to processing, both brine solution and brine ice were prepared. Salt (NaCl) was added and completely dissolved using tap water to each designated NaCl treatment solution by weight (w/w), and then placed in individual 20 gallon containers for carcass chilling. For Experiments I and II, 0, 4, and 8% NaCl brine solutions (w/w) were

prepared. Experiment III was prepared with 0, 1, 2, and 3% NaCl brine solutions (w/w), while Experiment IV contained 0, 3, and 4% NaCl brine solutions (w/w). One quart Ziploc bags were made for additional brine ice according to the target NaCl concentration. Both 20 gallon containers and Ziploc bags were placed overnight in a freezer room at -23°C (Figure 3.1).

Figure 3.1 Salt solution in Ziploc bags to be frozen.

3.2.2 Broiler Carcass Processing

For Experiments I and III, broiler birds were obtained from the Poultry Unit and processed in the Meat Processing Center at Cal Poly. A total of 15 male broilers (Ross 708, approximately 45 days old) were used for one replication (5 birds/treatment) in Experiment I, whereas a total of 48 male broilers (Ross 708, approximately 45 days old) were used for three replications (4 birds/treatment) in Experiment III. After 12 h feed withdrawal, birds were cooped and transported from the Cal Poly Poultry Unit to the Meat Processing Center. Each bird was shackled, electrically stunned for 3 s (40 mA, 60 Hz, 110 V) and bled for 90 s by severing both the carotid artery and jugular vein on one side of the neck. After bleeding, birds were subjected to scalding water (56°C) for 120 s, defeathered in a rotary drum picker (SP30 Ashley Sure-Pick, Ashley Machine Inc.,

Greensburg, IN) for 20 s, manually eviscerated, and washed. Resulting carcasses (4.8 pounds average weight) were hung on a shackle line and tagged on wing.

For Experiments II and IV, broiler carcasses (4.2 pounds average weight) were obtained from the broiler processing line at Foster Farms and chilled in the plant. In Experiment II, a total of 72 male broilers (Ross 708, approximately 45 days old) were used for 3 replications (24 birds/replication) conducted on three different days. In Experiment IV, a total of 48 male broilers (Ross 708, approximately 45 days old) were used for 4 replications (12 birds/replication) on four different days. Birds were randomly picked from the processing line at Foster Farms after evisceration and/or post-chilling.

3.2.3 Broiler Carcass Chilling

After evisceration and washing, broiler carcasses were chilled with mechanical agitation (VS-500, Grovhac Inc., Brookfield, WI) in a chilling solution (Figure 3.2). For Experiment I, broiler carcasses were chilled in one of three chilling treatments as follows: one control (0%NaCl/0.5°C) and two salt solutions (4% NaCl/-2.41°C and 8% NaCl/-5.08°C). For Experiment II, all birds were chilled by submersing in one of three chilling solutions as follows: one control (0% NaCl/0.5°C) and two salt solutions (4% NaCl/-2.41°C and 8% NaCl/-5.08°C) for chilling carcasses after evisceration or after pre-chilling in 0% NaCl/14°C or 0% NaCl/0.5°C. For Experiment III, all birds were chilled by submersing in one of four chilling solutions as follows: one control (0% NaCl/0.5°C) and three salt solutions (1% NaCl/-0.6°C, 2% NaCl/-1.2°C, and 3% NaCl/-1.8°C). For Experiment IV, broiler carcasses were chilled in one of three chilling solutions as follows: one control (0%NaCl/0.5°C) and two salt solutions (3% NaCl/-1.8°C and 4% NaCl/-2.41°C). Before chilling, one medium carcass per

chilling solution was selected for monitoring the internal breast temperature every 5 min until the carcass temperature reached ~ 4°C per USDA-FSIS regulations, using a digital thermometer logger (ThermaData Thermocouple Logger KTC, ThermoWorks, American Fork, UT). During chilling, sufficient ice or brine ice was added to maintain target solution temperatures. After chilling, carcasses were hung on a shackle and remained in the poultry cooler (1.1°C) until 3 h post mortem.

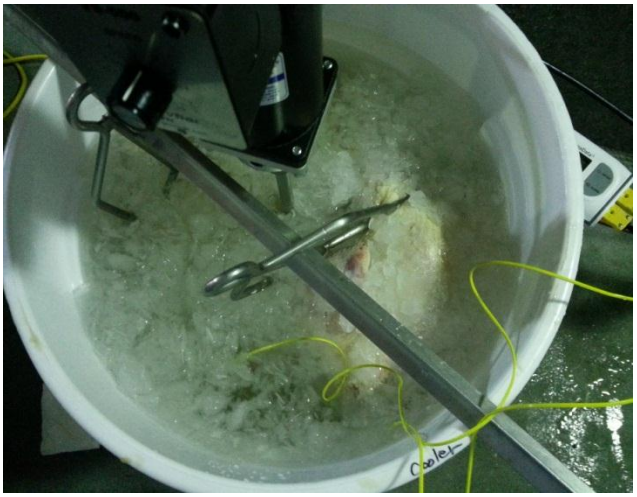

Figure 3.2 Broilers chilling in brine solution slurry with agitator.

3.2.4 Cooking Yield and Shear Force

Breast fillets were removed from each carcass 3 h post mortem (Figure 3.3). The right fillet of each carcass was placed into individual Ziploc bags and stored in the product cooler (2.2°C) for 24 h. The right fillets were weighed for a pre-cook weight. Fillets were placed on stainless trays on a stainless steel rack, covered with foil, and cooked to an internal temperature of 76.7°C in a convection oven (36S-Y1A Wolf Challenger XL Range, ITW Food Equipment Group LLC, Gleenview, IL) per USDA-Food Safety and Inspection Services (2001) guidelines. After cooking, the fillets were

reweighed (post-cook weight). The cooking yield was calculated as (post-cook weight)/(pre-cook weight) × 100.

Shear force was determined according to the razor-blade method described by Cavitt et al. (2004) using a texture analyzer (TAHDi, Texture Technologies Corp., Scarsdale, NY) calibrated with a 25-kg load cell. The razor blade (height, 24 mm; width, 8 mm) was set at 10 mm/s, and the test was triggered by a 10-g contact force. The shear force value (N) was calculated as the maximum force recorded during the shear. Two shear force measurements per breast fillet were made.

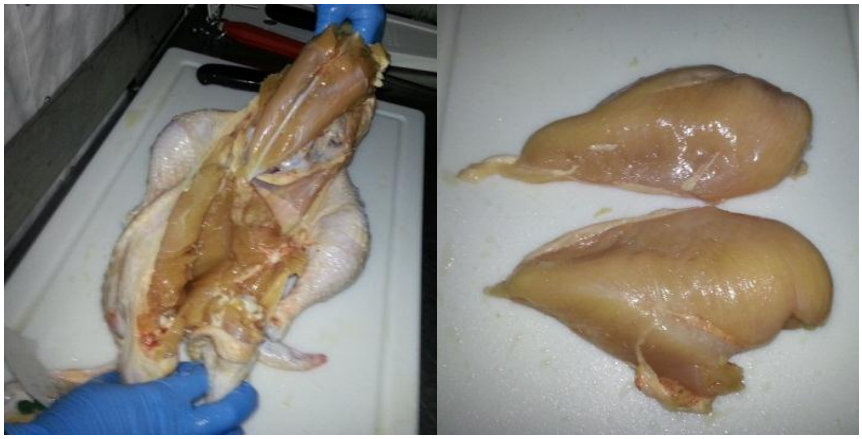

Figure 3.3 Removal of breast fillets from carcass and then labeled.

3.2.5 pH, R-value, and Sarcomere Length

After 3 h post mortem, breast fillets were removed from each carcass for Experiment III. The left breast was portioned into two cranial and one caudal piece (Figure 3.4). The resulting breast pieces were labeled, placed in Ziploc bags, and frozen in liquid nitrogen.

For pH measurement, cranial portion (2.5g) of the left breast fillet was pulverized and homogenized with 25 mL of a 5-mM iodoacetate solution with 150 mM potassium

chloride for 30 s using the method as described in Sams and Janky (1986). The pH of the homogenate was determined using a pH electrode (model 13 620 631, Fisher Scientific Inc., Houston, TX) attached to a pH meter (Accumet AR15, Fisher Scientific Inc., Pittsburgh, PA) calibrated at pH 4.0 and 7.0.

R-value (ratio of inosine:adenosine) was assessed as an indicator of adenosine triphosphate (ATP) depletion in the muscle using the method as described in Thompson (1987). Cranial portion (3 g) of the left breast fillet was pulverized and homogenized with 20 mL of 1M perchloric acid solution for 1 min then filtered through Fisher P8 filter paper. The amount (0.2 mL) of filtrate was added to 8.0 mL of 0.1M phosphate buffer to read an absorbance using a spectrophotometer (ThermoScientific GENESYS 10S UV-Vis Spectrophotometer, Fisher Scientific Co. LLC, Pittsburgh, PA) at 250 nm (IMP) and 260 nm (ATP). In comparison to the study by Kahn and Frey (1971) mentioned in Section 2.7.3, this experiment used IMP/AMP ratio to measure the R-value. Thus indicating that the higher the R-value, the more the ATP breakdown (or less ATP).

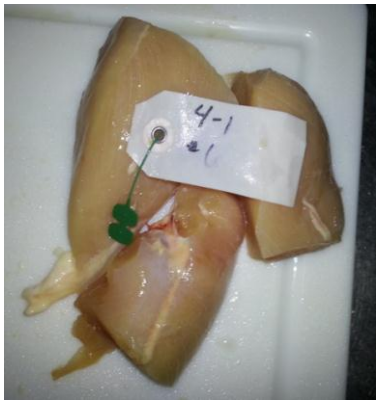

Figure 3.4 Left breast fillet cut into three portions to be evaluated for pH, R-value, and sarcomere length.

Sarcomere length was evaluated for the status of muscle contraction using a laser diffraction method (Cross et al., 1981). Caudal portion (10-15 g) of the left breast fillet

was homogenized in 50 mL solution composed of 0.25M sucrose, 2-mM potassium chloride, and 5-mM sodium iodoacetate. After homogenization, a drop of homogenate was placed on a slide with cover slip onto the laser platform where the slide will move until a diffraction pattern appears and was measured (Figure 3.5). The diffraction measurement was converted into sarcomere length using the equation:

$$Sarcomere\ Length\ = \frac{0.6328 * \mathrm{D} * \left[(\frac{T}{D})^2\ +\ 1\right]^{\frac{1}{2}}}{T}$$

0.6328 = Wavelength of the Helium-Neon laser light

D = Distance in mm from specimen to the diffraction screen

T = Distance in mm from the origin to the first order diffraction band

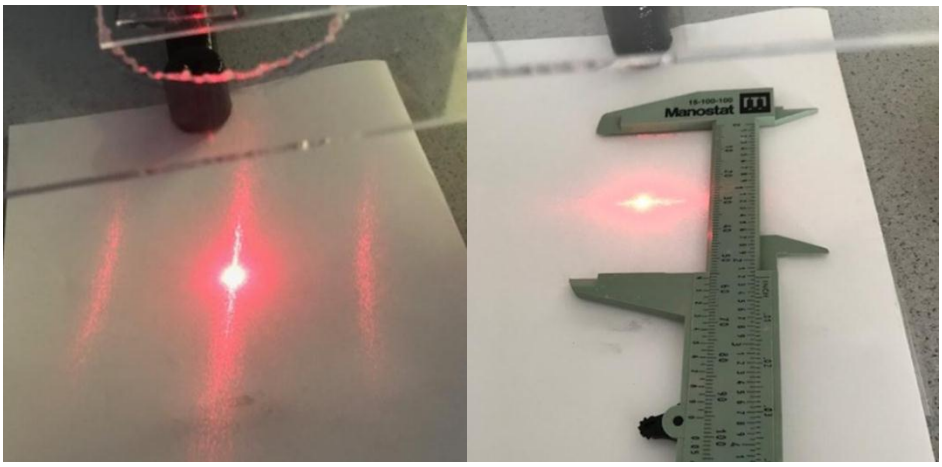

Figure 3.5 Laser diffraction pattern and band measurement technique for collecting sarcomere length.

3.2.6 Salt Content

For salt content analysis, breast samples of Experiment III were shipped and evaluated by Deibel Laboratories (Deibel Laboratories, S. San Francisco, CA). Cooked chicken breast sample (2.5–3 g) was moistened with 0.5M $AgNO_3$ solution and 15 mL

HNO_3, and then boiled until meat dissolved (~ 10 min). Concentrated aqueous $KMnO_4$ solution was added in small portions to the sample, boiled after each addition until $KMnO_4$ color disappeared and solution became colorless. After boiling, 25 mL H_2O was added and boiled for 5 min. The sample was then cooled, diluted to 150 mL, and mixed with 25mL ether by shaking. Once shaken, the sample was titrated with 0.1N NH_4SCN solution until solution became permanent light brown. After sampling, the difference was calculated by subtracting the mL 0.1M H_4SCN used from mL 0.1M $AgNO_3$ added. The calculated difference equals the amount of NaCl within the sample (AOAC, 2012).

3.2.7 Statistical Analysis

Experiments II and III were conducted with 3 replications and Experiment IV was with 4 replications. For all three experiments, data were analyzed by one-way ANOVA, using PASW 18 statistic program and a completely randomized design. A post-hoc analysis was performed using Tukey's HSD range test to evaluate differences among treatments ($P < 0.05$; SPSS, 2016).

3.3 Results

3.3.1 Experiment I: Carcass Chilling in 0%, 4%, and 8% Cold Brine Solutions

3.3.1.1 Shear Force

To compare the effects of saline chilling methods on breast tenderness, three salt concentrations and temperatures (0% NaCl/0.5^{o}C, 4% NaCl/-2.41^{o}C, and 8% NaCl/-5.08^{o}C) were initially used to chill carcasses. Control fillets in 0% NaCl showed a higher shear force than the fillets in brine chilling at sub-zero temperatures, regardless of salt content (Figure 3.6). The lower shear force values for breast fillets in

both 4 and 8% brine solutions indicates that the muscle is more tenderized than the fillets chilled in 0% NaCl.

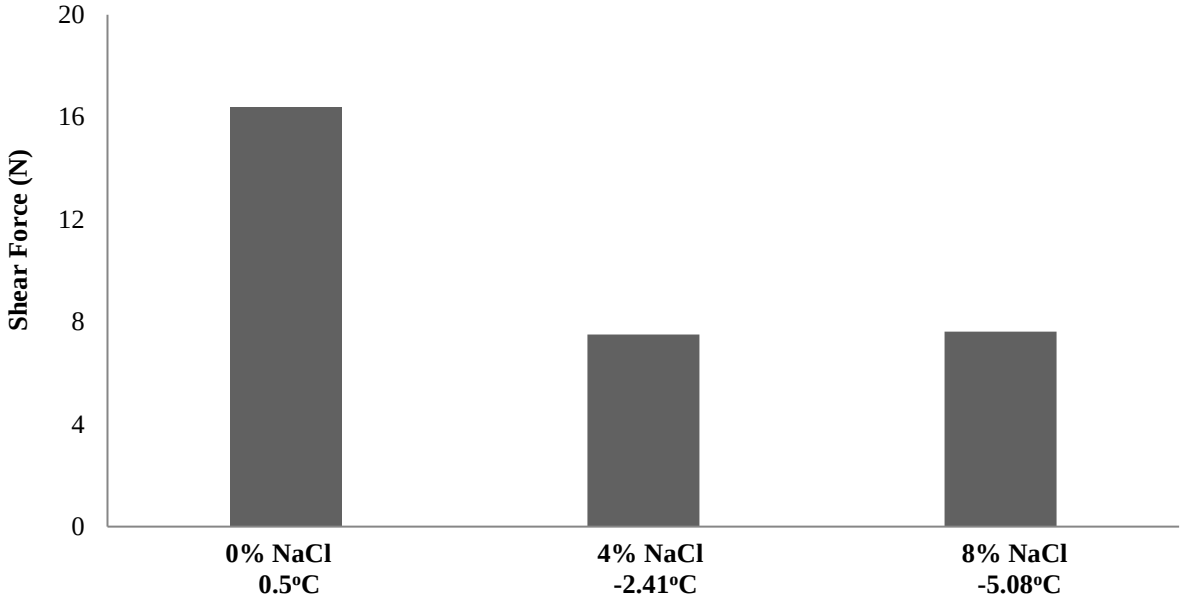

Figure 3.6 Effects carcass chilling[a] temperature and salt content on shear force of broiler breast fillets in Experiment I.
[a]0% NaCl/0.5ºC; Carcass chilling in ice slurry (0% NaCl) at 0.5ºC;
4% NaCl/-2.41ºC; Carcass chilling in 4% NaCl at -2.4ºC;
8% NaCl/-5.08ºC; Carcass chilling in 8% NaCl at -5.1ºC.

3.3.1.2 Cooking Yield

The cooking yield of breast fillets ranged from 75.7 to 81.8% with no significant difference, regardless of chilling method (Figure 3.7). Jeong et al. (2011) reported a similar cooking yield (75.1%) after water chilling for 57 min at 0°C, whereas a significant higher yield (78.3%) was found in 5% NaCl brine chilling overnight at -1°C than water chilling (73%) at 1°C after overnight chilling (Janky et al., 1978).

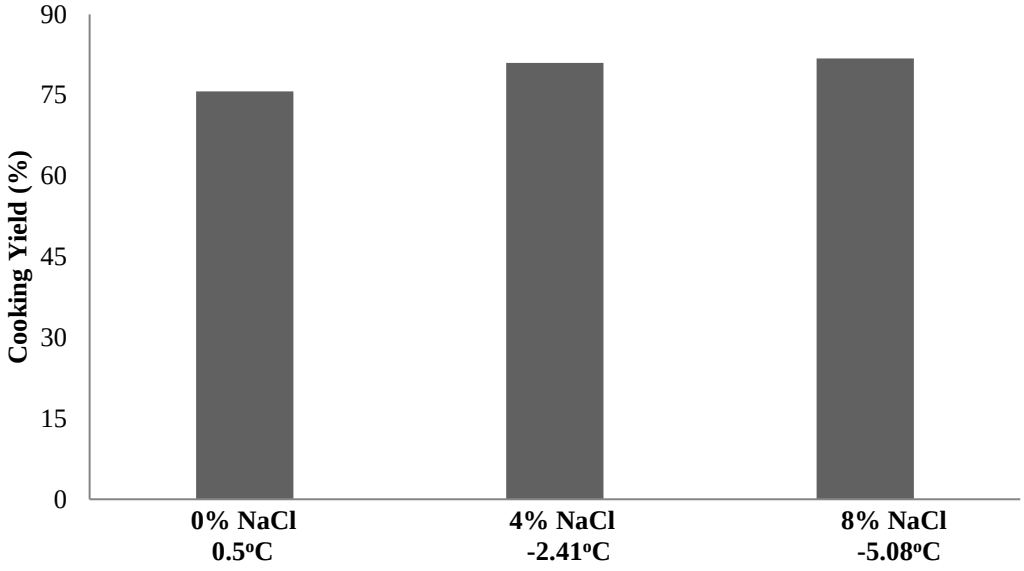

Figure 3.7 Effects of carcass chilling[a] temperature and salt content on cooking yield of broiler breast fillets in Experiment I.
[a]0% NaCl/0.5ºC; Carcass chilling in ice slurry (0% NaCl) at 0.5ºC;
4% NaCl/-2.41ºC; Carcass chilling in 4% NaCl at -2.4ºC;
8% NaCl/-5.08ºC; Carcass chilling in 8% NaCl at -5.1ºC.

3.3.2 Experiment II: Carcass Chilling in an Industry Chiller, 0%, 4%, and 8% Cold Brine Solutions

3.3.2.1 Shear Force

To compare various chilling effects on breast tenderness, three salt concentrations (0% NaCl/0.5ºC, 4% NaCl/-2.41ºC, and 8% NaCl/-5.08ºC) were used for carcass chilling with/without pre-chilling in 0% NaCl/0.5ºC or 0% NaCl/14°C. Fillets from the carcasses that were chilled in 4% NaCl/-2.41ºC significantly improved tenderness by reducing the shear force (12 – 14 N) of control to the level of 8.1 to 8.7 N, with no significant difference observed for the shear force (7.6 to 8.1 N) of 8% NaCl/-5.08ºC, regardless of pre-chilling (Figure 3.8).

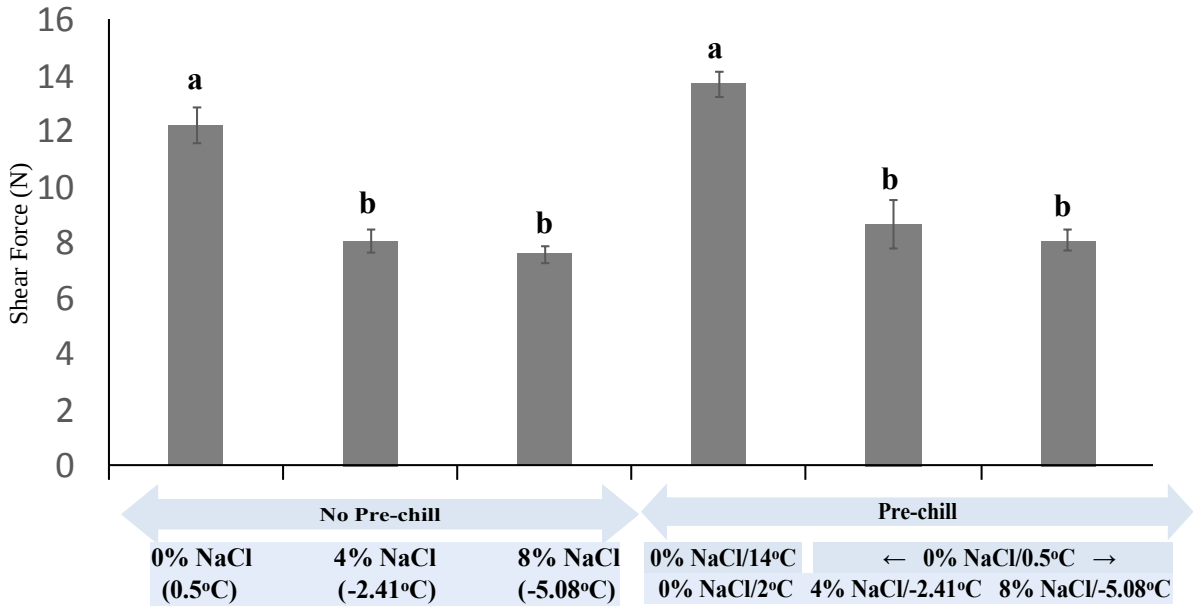

Figure 3.8 Effects of broiler chilling[a] temperature, salt content and chilling method on shear force of broiler breast fillets in Experiment II. Error bars represent ±SE; bars with differing letters (a,b) were significantly different ($P < 0.05$; n = 24).

[a]0% NaCl/0.5°C; Carcass chilling in ice slurry (0% NaCl) at 0.5°C;
4% NaCl/-2.41°C; Carcass chilling in 4% NaCl at -2.4°C;
8% NaCl/-5.08°C; Carcass chilling in 8% NaCl at -5.1°;
0% NaCl/0.5°C; Carcass chilling in 0% NaCl at 14°C (pre-chill) & 0% NaCl at 1.5 °C;
4% NaCl/-2.41°C; Carcass chilling in ice slurry (0% NaCl) at 0.5°C (pre-chill) & 4% NaCl at -2.4°C;
8% NaCl/-5.08°C; Carcass chilling in ice slurry (0% NaCl) at 0.5°C (pre-chill) & 8% NaCl at -5.1°.

3.3.3 Experiment III: Carcass Chilling in 0%, 1%, 2%, and 3% Cold Brine Solutions

3.3.3.1 Carcass Chilling

Before chilling, the internal temperature of eviscerated carcasses was ~ 40°C that was continuously reduced to 4.4 - 4.7°C during chilling, with average chilling times for 115, 100, 95, and 90 min in water control at 0.5°C and three brine solutions (1% NaCl/ -0.6°C, 2% NaCl/-1.2°C, and 3% NaCl/-1.8°C), respectively (Figure 3.9). Compared to water control, 3% NaCl/-1.8°C reduced the chilling time by 25 min (or 22%), with an intermediate reduction (13 - 17%) seen for other NaCl solutions.

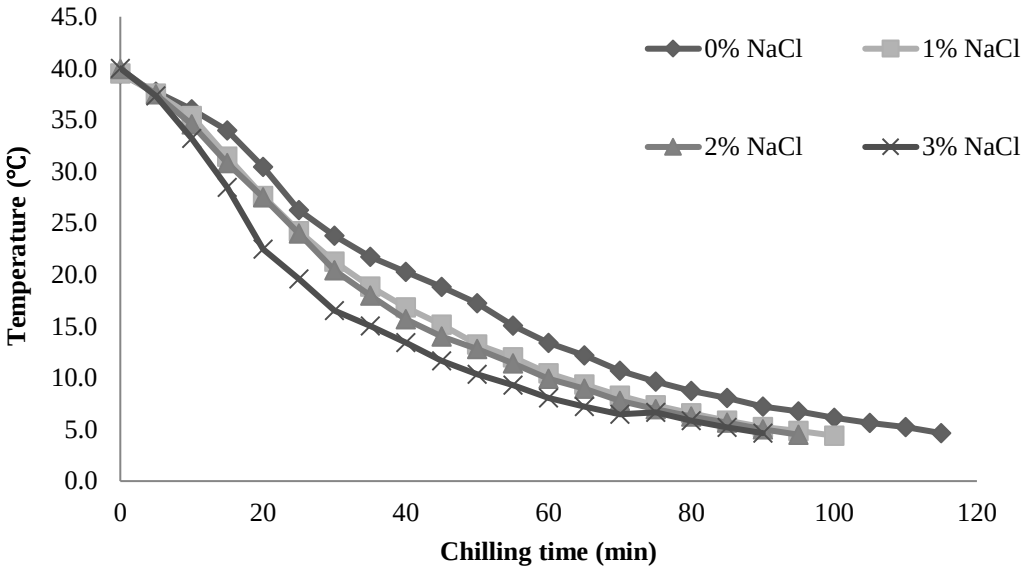

Figure 3.9 Temperature change profiles of broiler fillets during chilling[a] in water and brine solutions in Experiment III.
[a]0% NaCl/0.5ºC; Carcass chilling in ice slurry (0% NaCl) at 0.5ºC;
1% NaCl/-0.6ºC; Carcass chilling in 1% NaCl at -0.6ºC;
2% NaCl/-1.2ºC; Carcass chilling in 1% NaCl at -1.2ºC;
3% NaCl/-1.8ºC; Carcass chilling in 3% NaCl at -1.8ºC.

3.3.3.2 Carcass Chilling Yield, Fillet Salt Content, and Fillet Cooking Yield

After chilling, broiler carcasses were evaluated for weight gain that showed 2 – 3% increase with no significant difference, regardless of chilling method ($P < 0.05$) (Table 1). In evaluation of salt content in cooked skinless breast fillets, no significant difference was found between water chilling and brine chilling resulting in 0.06% ($P < 0.05$) (Table 1). The cooking yield of breast fillets ranged from 71.5 to 73.3% with no significant difference, regardless of chilling method ($P < 0.05$) (Table 1).

Table 1 Evaluation[1] of carcass chilling yield[2] and breast fillet property for salt content, pH, R-value, sarcomere length and cooking yield (±SE) after chilling carcasses in three different chilling solutions in Experiment III.

Chilling	0% NaCl/0.5°C	1% NaCl/-0.6°C	2% NaCl/-1.2°C	3%NaCl/-1.8°C
		Raw fillets		
Chilling yield (%)	$102^{a} \pm 0.44$	$102^{a} \pm 0.34$	$102^{a} \pm 0.20$	$103^{a} \pm 0.52$
pH	$5.64^{a} \pm 0.03$	$5.76^{a} \pm 0.05$	$5.70^{a} \pm 0.04$	$5.76^{a} \pm 0.04$
R-value	$1.37^{a} \pm 0.05$	$1.35^{a} \pm 0.05$	$1.36^{a} \pm 0.05$	$1.38^{a} \pm 0.04$
Sarcomere length (μm)	$1.29^{a} \pm 0.01$	$1.26^{a} \pm 0.02$	$1.25^{a} \pm 0.01$	$1.36^{a} \pm 0.09$
		Cooked fillets		
Cooking yield (%)	$73.3^{a} \pm 1.14$	$71.5^{a} \pm 1.02$	$72.9^{a} \pm 1.24$	$73.0^{a} \pm 0.85$
Salt content (%)	$0.06^{a} \pm 0.00$	$0.06^{a} \pm 0.01$	$0.06^{a} \pm 0.00$	$0.06^{a} \pm 0.01$

[a]Means within a row with unlike superscripts are different ($P < 0.05$).

[1]The number of observations in each chilling, n = 12.

[2]Same chilling conditions as in Figure 4.3

3.3.3.3 pH, R-value, and Sarcomere Length of Breast Fillets

After chilling, breast fillets measured pH 5.64 to 5.76 with no significant differences, regardless of chilling method ($P < 0.05$) (Table 1). Breast fillet R-values ranged from 1.35 to 1.38 with no significant differences, regardless of chilling method ($P < 0.05$) (Table 1). No R-value difference was reported between water chilling (R-value 1.37) and 3% NaCl brine chilling (R-value 1.38) at the time of boning (Table 1). After chilling, breast sarcomere lengths were measured with no significant differences, regardless of chilling method ($P < 0.05$) (Table 1). There was no length difference between water chilling (1.29 μm) and 3% NaCl brine chilling (1.36 μm) at the time of boning (Table 1).

3.3.3.4 Shear Force

The shear force of cooked breast fillets was measured using the method of razor-blade (Cavitt et al., 2004). The shear force decreased from 16.1 to 11.5 (N) in a step wise pattern as the salt content increased and solution temperature decreased from 0%/0.5°C to

3%/-1.8°C, with the lowest shear force (most tenderization) observed in 3% NaCl brine at -1.8°C ($P < 0.05$) (Figure 3.10). Comparing the results of Experiment II and III, we expect that the maximal tenderness in breast fillets could be achieved when carcasses are chilled in 3 or 4% NaCl brine solutions at subzero temperatures.

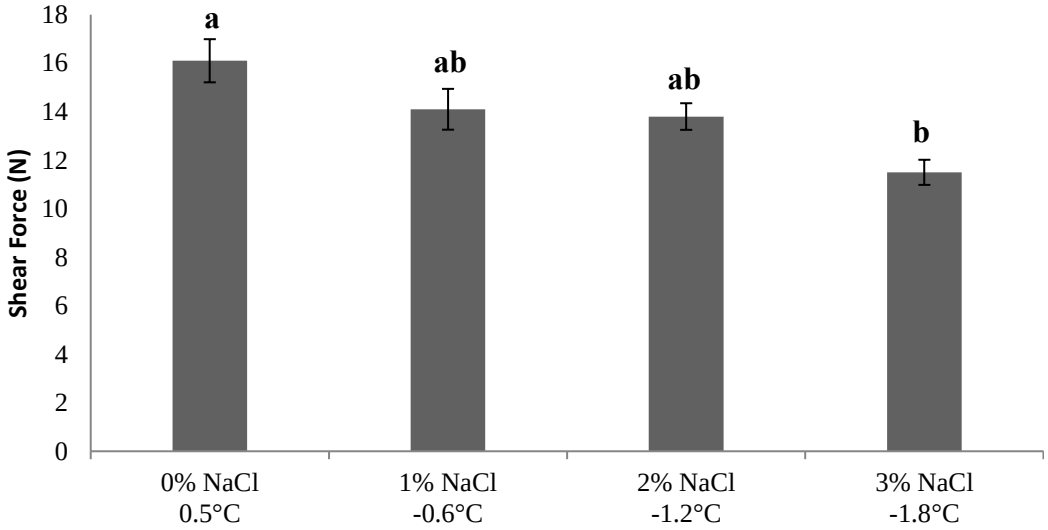

Figure 3.10 Effects of broiler chilling[a] temperature and salt content on shear force of broiler breast fillets in Experiment III. Error bars represent ±SE; bars with differing letters (a,b) were significantly different ($P < 0.05$; n = 12).
[a]0% NaCl/0.5ºC; Carcass chilling in ice slurry (0% NaCl) at 0.5ºC;
1% NaCl/-0.6ºC; Carcass chilling in 1% NaCl at -0.6ºC;
2% NaCl/-1.2ºC; Carcass chilling in 1% NaCl at -1.2ºC;
3% NaCl/-1.8ºC; Carcass chilling in 3% NaCl at -1.8ºC.

3.3.4 Experiment IV: Carcass Chilling in 0%, 3%, and 4% Cold Brine Solutions

3.3.4.1 Broiler Carcass Chilling

Before chilling, the internal temperature of eviscerated carcasses was ~40°C that was continuously reduced to 4.4 - 4.7°C during chilling, with average chilling times of 90, 80, and 55 min in water control at 0.5°C and two brine solutions (3% NaCl/-1.8°C and 4%NaCl/-2.4°C), respectively (Figure 3.11). Compared to water control, 4% NaCl reduced the chilling time by 35 min (or 39%), with an intermediate reduction (11%) seen for 3% NaCl solution. The chilling time (80 min) in the 3% brine solution in this experiment was similar to the chilling time (90 min) of the 3% solution in Experiment III.

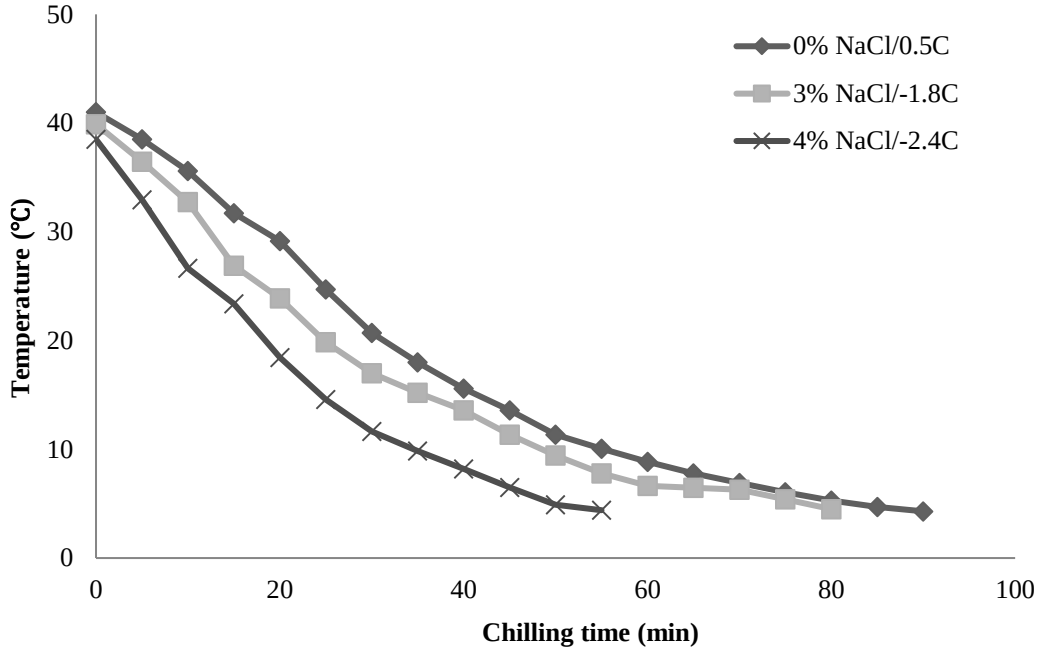

Figure 3.11 Temperature change profiles of broiler breast fillets during chilling[a] in water and brine solutions in Experiment IV.

[a]0% NaCl/0.5°C; Carcass chilling in ice slurry (0% NaCl) at 0.5°C.

3% NaCl/-1.8°C; Carcass chilling in 3% NaCl at -1.8°C.

4% NaCl/-2.4°C; Carcass chilling in 4% NaCl at -1.8°C.

3.3.4.2 Shear Force

To identify the best chilling condition based on Experiments I, II, and III, three salt concentrations (0% NaCl/0.5ºC, 3% NaCl/-1.8 ºC, and 4% NaCl/-2.41ºC) were selected and evaluated for broiler carcass chilling. Control fillets in 0% NaCl/0.5ºC showed a higher shear force (12.64 N) than the fillets in 4% NaCl/-2.41ºC (8.4 N) ($P < 0.05$), with an intermediate value of 10.15 N in 3% NaCl/-1.8 ºC (Figure 3.12). Results indicated that shear force reduced in a stepwise pattern as the NaCl increased from 0 to 4% in the chilling solution and chilling temperature was reduced from 0.5 to -2.41ºC (Figure 3.12). Comparing the previous results to Experiment IV, we expect that an ideal chilling condition for broiler tenderization and practical application would be 4% NaCl solution at -2.41ºC.

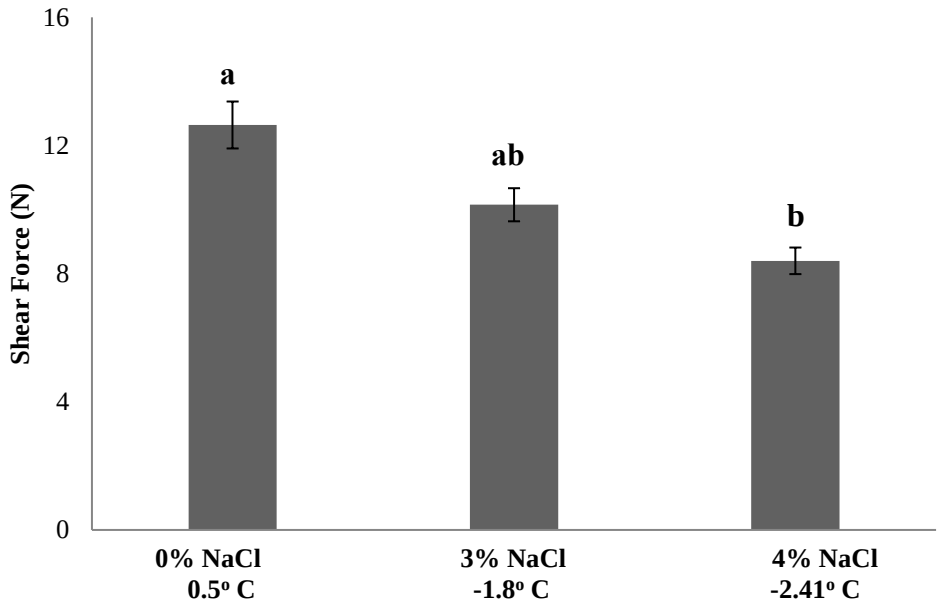

Figure 3.12 Effects of broiler chilling[a] temperature and salt content on shear force of broiler breast fillets in Experiment IV. Error bars represent ±SE; bars with differing letters (a,b) were significantly different ($P < 0.05$; n = 16).
[a]0% NaCl/0.5ºC; Carcass chilling in ice slurry (0% NaCl) at 0.5ºC.
3% NaCl/-1.8ºC; Carcass chilling in 3% NaCl at -1.8ºC.
4% NaCl/-2.4ºC; Carcass chilling in 4% NaCl at -2.4ºC.

3.4 Discussion

Based on the results of this study, there is a trend of shear force reduction as the salt concentration increased and the chilling temperature decreased in the chilling solutions. The shear force of the breast fillets noticeably decreased in a continuous manner once the salt concentration exceeded 2% NaCl and temperatures decreased below -1.2 ºC (Figure 3.13). Although improved tenderness of breast fillets required a brine chilling solution of at least 3% NaCl, the 4% NaCl treatment may be the most ideal in terms of improved shear force without the need of using the 8% NaCl solution. Duke and Janky (1984) used one water control and 27 treatment combinations using three brine chilling solutions (2.5, 5.0 and 7.5% NaCl), three chilling temperatures (21, 13, 1°C) and three chilling steps (15 min/21°C, 15 min/13°C, and 15 min/1°C). Out of the 28 chilling

treatments, the two lowest shear forces were found in the combination of 5% NaCl brine for 45 min in three combinations (15 min at 21°C, 15 min at 13°C and 15 min at 1°C) and 2.5/5.0/7.5% NaCl for 15 minutes each at 21°C /13°C /1°C, respectively. Whereas the highest shear force was observed in the water control of 0% NaCl for for 45 min in three combinations (15 min at 21°C, 15 min at 13°C and 15 min at 1°C). Comparing water chilling and 5% NaCl brine chilling for 60 min (30 min at 21°C and 30 min at 1°C), Sams and Janky (1986) reported no tenderness difference with significantly higher sodium content found after the brine chilling.

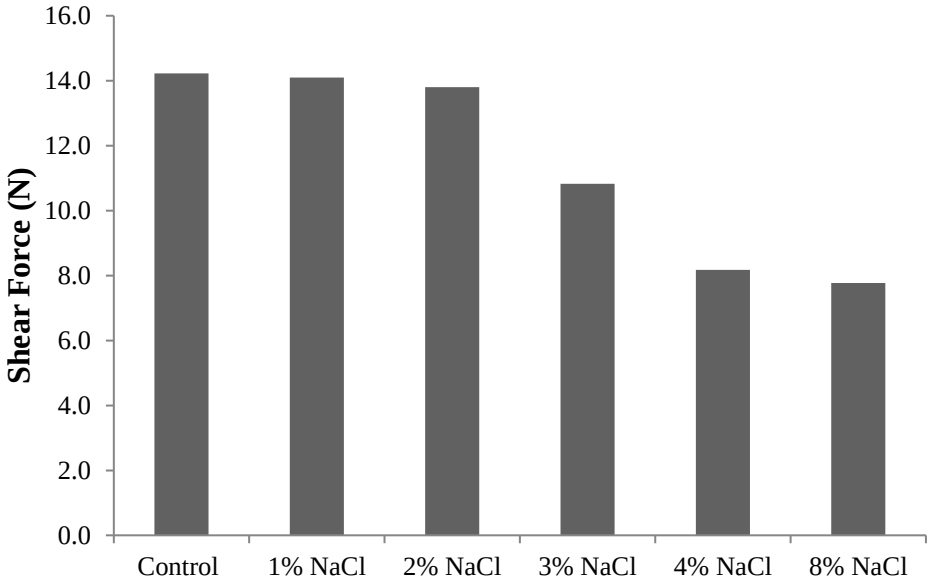

Figure 3.13 Shear force averages for each brine solution chilling treatment[a] across Experiments I, II, III, and IV.
[a]0% NaCl/0.5ºC; Carcass chilling in ice slurry (0% NaCl) at 0.5ºC;
1% NaCl/-0.6oC; Carcass chilling in 1% NaCl at -0.6 ºC;
2% NaCl/-1.2oC; Carcass chilling in 1% NaCl at -1.2 ºC;
3% NaCl/-1.8ºC; Carcass chilling in 3% NaCl at -1.8ºC;
4% NaCl/-2.4ºC; Carcass chilling in 4% NaCl at -2.4ºC.

Previously, the amount of 0.2 - 0.76% NaCl was reported in cooked fillets when broilers were chilled in 5% NaCl brine for 4 h at -1°C or for 45 min in three combinations (15 min at 21°C, 15 min at 13°C and 15 min at 1°C) (Dukes and Janky,

1984; Hoey et al., 1983). Part of the reason for the lower salt content in our study than the previous is expected from the shorter chilling time (115 vs 240 min) and/or sub-zero chilling temperature (-0.6 to -1.8°C vs 1 to 21°C), with no pre-chilling step. According to USDA National Nutrition Database (2016), the average salt in skinless raw chicken breast is 45 mg sodium/100g. Jeong et al. (2011) reported a similar cooking yield (75.1%) after water chilling for 57 min at 0°C, whereas a significant higher yield (78.3%) was found in 5% NaCl brine chilling at -1°C than water chilling (73%) at 1°C after overnight chilling (Janky et al., 1978).

Although the tenderness appears to be improved as the salt concentration increased within chilling solutions, there was no supporting data as to what mechanism was responsible for such results when comparing physicochemical analysis up to 3% NaCl chilling (Table 1). These results may be due to the small sample size tested within Experiment III or the less sufficient salt content with 3% NaCl chilling. However, the results in this study were found to be similar to the breast pH values (pH 6.14), R-values (R-value 0.99), and muscle sarcomere lengths(1.64 μm) reported after water chilling for 62 min at 0.2°C by Sansawat et al. (2014). Although no significant difference was found with 3% NaCl chilling for meat qualilty improvement factors, except for meat tenderness, some of these factors could be detected and supportive when the breast fillets from 4% and 8% NaCl brine chillings are evaluated for pH, R-value, sarcomere length, and cooking yield.

In general, chilling broilers in sub-zero saline solutions has the potential to improve processing productivity and product quality. Using sub-zero saline solutions for this study, the chilling time decreased as the salt percentage of the treatment increased

and chilling temperature decreased. This could potentially increase the number of broilers that are processed by sending more carcasses through the chiller. With faster chilling rates, this could also reduce the amount of water required for processing and wastewater produced. Furthermore, as the salt percentage increased and chilling temperature decreased, the product quality improved in terms of breast fillet tenderness. Based on these results, increasing the salt concentration to maintain low sub-zero temperatures within the chiller can provide advantages in processing efficiency, production productivity, and product quality.

Chapter 4
CONCLUSIONS

Due to the high demand for water consumption and increased processing costs in poultry processing plants, the improvement of poultry chilling efficiency and product quality is desirable for poultry processors and consumers. In this study, broiler carcasses have been chilled using various saline solutions and chilling temperatures from 0 to 8% salt and from 0.5 to -5.08°C. Broiler chilling efficiency and breast fillet tenderness were significantly improved as the chilling temperature reduced from 0.5 to -5.08°C, and the salt content increased from 0 to 8%, with no significant difference observed for 4% NaCl/-2.41°C and 8% NaCl/-5.08°C. Based on these results, chilling of broiler carcasses in 4% NaCl/-2.41°C appears to be ideal to improve chilling efficiency and meat tenderness compared to the control chilling in 0% NaCl/0.5°C, with the potential opportunity for water saving and product safety improvement.

Chapter 5

AREAS FOR FURTHER STUDY

The following topics are recommended for future study:

- Microbiological populations on broiler carcasses before and after sub-zero brine solution chilling.
- Sensory analysis of finished products.
- Analysis of water saving, waste management, and overall processing cost.
- Analysis of raw meat quality of broilers chilled in 4%NaCl/-2.41ºC and 8% NaCl/-5.08ºC.
- Implementation of sub-zero brine solution chilling to turkey in order to improve chilling efficiency, product quality, and microbial safety.

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