SAVORPHOS AS AN ALL NATURAL PHOSPHATE REPLACER IN WATER AND OIL BASED MARINADES FOR ROTISSERIE BIRDS AND BONELESS-SKINLESS BREAST

A Thesis

by

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As consumer demand for all-natural marinades increases, the need to replace phosphate with a natural product that can produce equivalent or improved yield in products such as but not limited to rotisserie chickens (RWOG) and boneless/skinless breast (BSB) is a challenge for processors. The objective of this study was to determine if using an all-natural non-phosphate blend (savorphos-200, SP) in water-based (WB) and oil-based (OB) marinades would perform better in quality and yield parameters than a commercial phosphate blend (PB). The treatments included WB+PB (water, 0.4% phosphate, 0.7% salt), WB+SP (water, 0.5% savorphos-200, 0.7% salt), OB+PB (water, 3% canola oil, 0.4% phosphate, 0.7% salt), and OB+SP (water, 3% oil, 0.5% savorphos-200, 0.7% salt).

RWOG and BSB were injected with a multi-needle injector to 20% (wt/wt) pick-up at a constant pressure (15-20 psi). The parameters measured were marinade pick-up %, 20 min and 24 hr marinade retention %, and cook loss %. Color, tenderness, total moisture, and sensory test were conducted on BSB. Data were analyzed within marination type (WB and OB). Results for the RWOG indicated SP obtained higher
pick-up yield (p<0.05) and lower cook loss in the OB marinade compared to the PB. For the BSB, pick-up yield on OB marinades are higher for SavoPhos (p<0.05) when compared to the PB. On WB marinades cook loss was lower on SavorPhos compared to the PB. On RWOG and BSB variability is lower for SP on pick-up and 20 min retention yield values. Texture shear values were lower (p<0.05) on SP samples when compared to the PB. A consumer triangle sensory test was not able to distinguish between treatments (p>0.05). Therefore, savorphos-200 can be used as a natural non-phosphate blend in water based marinades with no detriment to yield. In addition, savorphos-200 can be used as a natural non-phosphate blend in oil-based marinades with yield improvements.
I would like to thank all the members of our amazing lab team, Guilberto Veluz, Ana Sanchez, Micheal Yeater, and Kurt Zihlman for their continuous and unconditional help running samples as well as for their friendship.

I would like to thank my committee members, Dr. Craig Coufal, Dr. Matthew Taylor, and Dr. Cain Cavitt for their help in the elaboration of this thesis and professional guidance.

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<tr>
<td>RWOG</td>
<td>Rotisserie Whole Bird Without Giblets</td>
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<td>WHC</td>
<td>Water Holding Capacity</td>
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<td>BSB</td>
<td>Boneless/Skinless Breast fillets</td>
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<td>hr.</td>
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<td>w/w</td>
<td>Weight over Weight</td>
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<td>FSIS</td>
<td>USDA Food Safety and Inspection Service</td>
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<tr>
<td>ATP</td>
<td>Adenosine Tri-Phosphate</td>
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<tr>
<td>CIE</td>
<td>International Commission on Illumination</td>
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<td>STPP</td>
<td>Sodium Tri-PolyPhosphate</td>
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<td>pH</td>
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Marination of whole birds and boneless/skinless breast fillets is a growing trend. Consumers seek the convenience, versatility, and quality obtained from marinated products (Owens et al., 2009). Poultry meat marinated with water, salt, and phosphate is the most common form of enhancement of meat products for the foodservice and retail markets (Heath and Owens, 1987; Alvarado and McKee, 2007; Petracci et al., 2012).

A growing market for natural products demands ingredients that can be labeled as All Natural. Under current FSIS regulation phosphates are not considered a natural ingredient. Processors need to find functional ingredients that can perform similarly to phosphate’s water holding capacity (Fernandez-Gines et al., 2004).

Gums, fibers, and non-modified starches have been used and tested as phosphate replacers. All the latter have a product that can be labeled as All Natural. When used at manufacturer recommended amounts similar water holding capacity as phosphates are obtained without affecting sensorial or quality attributes negatively (Keeton et al., 2003; Fernandez-Gines et al., 2004; Aleson-Carbonell et al., 2005).

SavorPhos is a proprietary blend of all natural flavorings, citrus flour, and less than two percent of sodium carbonate as a processing aid that is currently being used as an all-natural ingredient for chicken marinades.
The high fiber and low protein content are believed to contribute to the water
holding capacity of SavorPhos (Marin et al., 2007).

The objectives of this study were to test the hypothesis that SavorPhos can act as
an all-natural phosphate replacer and to compare the performance of SavorPhos AF-200
against the commercially available phosphate blend on the quality and acceptability of
the rotisserie chicken and boneless/skinless chicken breast using water and oil-water
based marinades.
CHAPTER II

LITERATURE REVIEW

Rotisserie Birds and Boneless/Skinless Breast Fillets

The U.S.A. yearly per capita consumption of broiler meat reached 82.9 pounds in 2011, with an estimated 46% of this production marketed and sold as further processed poultry products (NCC, 2012). Further processed poultry products include but are not limited to marinated rotisserie chicken and boneless chicken meat sold as either ready-to-cook (RTC) or ready-to-eat (RTE) products. Rotisserie birds (RWOG) sold to the foodservice industry are commonly enhanced to improve sensory attributes (tenderness and juiciness) for consumers and to compensate for the water lost during cooking. Boneless/skinless breast fillets sold to either foodservice or to retail stores are also enhanced to improve yields and sensory attributes (tenderness and juiciness) (Alvarado and McKee, 2007; Owens et al., 2009).

These two types of poultry products present different challenges during processing. The RWOG is a bone-in product that is composed of white and dark meat with skin, while the boneless/skinless breast fillet is entirely white meat with no skin. The white meat portion of the whole birds is composed of the breast fillets, the tender, and the wings, while the dark meat portion includes the thighs and the drums (Owens et al., 2009). The difference of color in the meat is attributed to the amount of myoglobin present in the muscle (Davis and Franks, 1995). Myoglobin is a globular protein that
contains a heme group that produces a distinctive red color when oxygen is bound, and allows the transport of oxygen through the muscle (Kranen et al., 1999; Owens et al., 2009). Higher activity muscles, such as thighs and drums, require more oxygen. White meat has lower fat (1.8% vs. 9.3%), connective tissue (10.0% vs. 11.9%), and higher protein content (86.3% vs. 71.7%) when compared to dark meat w/w (Rizzi and Chiericato, 2010). The boneless/skinless breast fillets are manually or automatically extracted from the whole bird.

Under current labeling laws (9 C.F.R. §381.169), marinated products must contain eight percent or more of a solution, and under the product name use the labels “enhanced with,” “may contain up to,” or other statements approved by the law and its amendments. Poultry companies used marination or enhancement of poultry meat as early as the 1950s, providing consumers and the foodservice industry with poultry products that are more desirable and palatable (Owens et al., 2009). Recently, marinated products have increased in foodservice and retail as consumers demand more versatility, convenience, and variety (Pollock, 1997). Enhancement also provides an economic benefit to processors by allowing an increase in processing yields (Alvarado and McKee, 2007).

**Meat Quality**

The muscle is a combination of water, proteins, carbohydrates, and fats. The concentration of fats and proteins, and the types of proteins present vary depending on the muscle origin (Owens et al., 2009; Rizzi and Chiericato, 2010). Actin and myosin are the most abundant and functional of all the proteins present in the muscle (Alvarado and
McKee, 2007). For muscle to be considered meat it has to undergo the biochemical process of rigor mortis. As blood flow is removed through the exsanguination process during slaughter of chickens, oxygen supply is terminated, limiting the amount of ATP generated through glycolysis, causing the muscle to start anaerobic respiration. Phosphorylation of ADP does not occur at the same rate as usage of ATP (Young and Lyon, 1997a). The decrease of ATP availability limits the ability of the calcium pumps to extract calcium from the muscle filaments. The presence of calcium activates the tropomyosin complex to expose the binding site in the globular protein actin to which the myosin head binds and forms the actomyosin bond. In normal conditions, the removal of calcium and the presence of ATP break the actomyosin bond, thus achieving relaxation of the muscle (Young and Lyon, 1997a). Under rigor mortis, these bonds form and cause the stiffening of the muscle (Xiong, 2004). Anaerobic respiration increases the amount of lactic acid in the muscle through the Krebs cycle, which decreases muscle pH from the normal value of ~7.2 to 5.7-6.0. This decrease in pH is believed to weaken the myosin head, which, together with the strength of the contraction forces being generated results in the cleavage of the actomyosin bond, and some degree of relaxation is achieved. It is at this point of relaxation and a pH of 5.7-6.0 that rigor mortis is considered to have resolved, and muscle can be called meat (Sams, 1990; Owens et al., 2009).

Meat quality can be affected by several factors, including color, water holding capacity (WHC), pH, and texture parameters of the meat (Alvarado and McKee, 2007;
Gorsuch and Alvarado, 2010). The relative contributions to meat quality of these factors are reviewed here.

**Color**

Color is measured according to the CIE L*a*b* scale (CIELAB) where L* represents the lightness, a* the redness, and b* the yellowness of an object. The CIELAB scale was introduced to limit the variation between sample measurements via the quadratic values of the CIE Tristimulus values (Mancini and Hunt, 2005). Color variation of meat is of great importance to the consumer acceptability and meat functionality (Alvarado and McKee, 2007; Owens et al., 2009; Gorsuch and Alvarado, 2010). Consumers become concerned when boneless/skinless breast meat is too pale or too dark (Barbut, 2009). Wilkins et al. (2000) reported that color extremes are likely to be discriminated at the point of purchase. In meat, color can be affected by the amount and state of myoglobin, pH, and whether the meat is marinated or not. Dark meat contains more myoglobin, which results in a higher red value and lower lightness on the scale. Higher fat content, contained in specific areas of the thigh, may possibly increase the L* values. The color of myoglobin depends on the oxidation state of the heme group (Kranen et al., 1999; Owens et al., 2009). When oxygen is bound, a bright red color is generated. Meat pH values have been correlated to color measurements (L*- lightness value) of boneless/skinless breast, where the lighter the color is, the lower the pH (Alvarado and Sams, 2004; Alvarado and McKee, 2007; Barbut, 2009; Owens et al., 2009). Marinated chicken breasts have been reported to result in higher L* values and lower a* and b* when compared to non-marinated meat (Barbut, 2009; Gorsuch and
Alvarado, 2010). These changes were attributed to the increase of free water present between the myofibrils, causing the light from the colorimeter to be reflected in different directions.

**Water Holding Capacity**

Water holding capacity is the amount of water that a meat product can retain within the muscle fibers through capillary and ionic force (Alvarado and McKee, 2007). In meat, water is present in three forms: bound (4-5%), immobilized (35-37%), and free water. Free water is easily lost over time as it is held between the fibers mainly by capillary forces. Immobilized water is the result of electrical charges that form electrical bonds between the water molecules and the proteins present between the muscle fibers. Bound water is all the water bound chemically in the meat structure and cannot be freed by any means (Hedrick et al., 1989; Honikel and Hamm, 1994). The ability to hold the water can be gravely affected by the final pH of the meat (Alvarado and McKee, 2007; Petracci et al., 2012). Research indicates that as pH increases closer to neutrality, the amount of ionic charges increase between the muscle fibers, thus increasing the amount of water moved from free state to immobilized, also increasing the amount of water that can be retained. As pH approaches the iso-electric point (pI) of 5.1 of actin and myosin (Heath and Owens, 1987; Owens et al., 2009) the electrical charge of the protein fibers nets zero, and the water holding capacity of free water is lost, thus causing a pale and exudative appearance of meat. These two effects can greatly affect texture (Heath and Owens, 1987; Alvarado and McKee, 2007; Gorsuch and Alvarado, 2010). The higher the meat pH is, the higher the WHC of meat and better is the texture perception of the
cooked meat by consumers, due to the higher water content. At a low pH, consumers perceive a drier and harder cooked product. Young and Buhr (2000) reported an increase of 0.12 pH units when marinating chicken with STPP and salt, compared to just salt 0.03 pH, resulting in lower cook loss (15.4% vs. 16.9%) and shear values (5.9kg vs. 6.1kg). Marination has been used as a method to increase the content of water in the meat, as well as to move the pH closer to 7.0 by using highly alkaline ingredients and increasing the WHC of meat (Barbanti and Pasquini, 2004). Along with water, common ingredients used in marinades include salts and phosphates (Alvarado and McKee, 2007; Saha et al., 2009). Foodservice may even include oils in the marinade to boost palatability of lean cuts.

**Texture**

Texture can be defined as the manifestation of the composition of a product through their reaction to stress, moisture content, and tactile feel (Meilgaard et al., 2007). The enhancement of poultry products with water and oil-water based marinades provides consumers with a product that is juicier, more tender, and more flavorful (Post and Heath, 1983; Alvarado and McKee, 2007). Texture is an attribute that impacts the perceived quality of meat by consumers (Owens et al., 2009; Saha et al., 2009). The increase in demand of poultry has led to the harvesting of the muscles before the onset of rigor has been completed (Alvarado and Sams, 2004; Alvarado and McKee, 2007). This generates a tougher poultry product on the market. Therefore, marination has been used on early, deboned chicken to increase the water content thus improving texture and sensorial values.
Texture can be evaluated with precise mechanical apparati like the Instron universal testing machine. Shear force values are obtained with the allo-kramer testing head. The method measures the required force used to shear a sample of breast meat in a perpendicular alignment from the direction of the meat fibers. Data obtained from the allo-kramer shear force method need to be anchored with consumer sensorial tests to assess the relevance of these values with regard to texture (Smith et al., 1988; Owens et al., 2009).

Marination

The main purpose of marination is to improve the sensory experience of the consumer by improving tenderness, juiciness, flavor, and appearance (Young and Buhr, 2000; Barbanti and Pasquini, 2004; Alvarado and McKee, 2007). Consequently processors obtain better yields and consumers obtain better quality products that are more convenient, e.g. pre-marinated individually frozen chicken that is RTC or RTE microwavable marinated chicken. (Alvarado and McKee, 2007). Retail markets mostly use water-based marinades due to customer concern over label and nutritional statements. Oil-water marinades are more common in the foodservice and convenience food market than in retail (Cunningham and Tiede, 1981). Higher contents of oils increase desirability of meat products to consumers due to their ability to improve mouth-feel and juiciness, thus incentivizing the foodservice industry to include oils in their formulations (Heath and Owens, 1987; Yackel and Cox, 1992; Smaoui et al., 2012). Labeling concerns have led the industry to remove high caloric content oils, and develop “cleaner labels” (e.g. enhanced with chicken broth) containing other all-natural
ingredients (Saha et al., 2009; Morey et al., 2012). Labeling laws combined with consumer trends and new functional ingredients have played an important role in regards to the formulation of the marinade, the type of marination method to be applied, and uptake yield target of marinated meat products (FSIS, 2005; Saha et al., 2009). The type of product (rotisserie birds or boneless/skinless breast fillets) dictates different constraints in the method of introduction of the marinade. Surface area, functional proteins, target yield, ingredients, and presence of fat, all play a role in attaining the desired yields (Alvarado and Sams, 2004; Alvarado and McKee, 2007; Owens et al., 2009; Bowker et al., 2010b). Greater surface area of the meat exposed to the marinade allows for more contact to the functional proteins (myosin and actin) which increases the WHC. The amount of fat will interfere with the WHC of a muscle as it reduces the amount of functional protein (Rizzi and Chiericato, 2010). Marination methods exist that permit the optimization of the marinade uptake.

Methods of Marination

Several methods are in place to achieve the desired uptake, yet all have different strengths and weaknesses. The oldest marination method known is the soak method (Owens et al., 2009). Salt and water brine is prepared and the whole meat portions are allowed to soak over a period of 24-48 hr under refrigeration. This method is very dependent on surface area and osmosis. The higher salt concentration of the brine transfers to the meat over time until equilibrium between marinade and meat is reached. During this process water is pulled *via* osmosis into the muscle. Penetration is limited to
the surface contact areas and is time-dependent (Alvarado and McKee, 2007; Owens et al., 2009).

Injection is another method of introducing marinade into a meat block. The process consists of forcing a marinade with pressure through a needle with a single or multiple holes (Alvarado and McKee, 2007; Owens et al., 2009). The injection process can be done by hand with a syringe, with a single or multiple needles, or automatically with a multiple needle injector. The latter is preferred for bone-in products, and possesses the advantage of constant pressure, which permits equal dispersal of the marinade within the meat, thus preventing the formation of marinade pockets. Yield targets are also easier to meet and can reach high levels of injection. The drawback of using the needle method is that physical punctures are left behind on the meat surface. Needle size and needle pores limit the use of spices or other functional ingredients, as clogging can occur and disrupt the injection process (Post and Heath, 1983; Alvarado and McKee, 2007; Gorsuch and Alvarado, 2010; Petracci et al., 2012). The puncture holes are believed to allow the purge (loss of brine) of water, as they act as channels through which brine escapes. To help counteract the needle marks, processors use a combination of methods, where the meat is first injected and then tumbled marinated.

Tumble marination is the use of vacuum and a massaging effect to force water into the muscle. Vacuum draws air present between the meat fibers. A swelling effect is also caused when vacuum is applied, allowing the penetration of the brine within the muscle. Together with the massaging effect and the brine, the salt-soluble actin and myosin proteins are extracted, allowing for an increase in WHC of the meat (Alvarado
and McKee, 2007). As extraction increases, more protein binding sites are exposed, permitting protein-water interaction to occur (Bowker et al., 2010b). Unlike injection, tumbling is limited to the surface area of the whole meat portion in contact with the brine. Higher pick-up target requires extended tumbling periods, which can cause over extraction of proteins (Heath and Owens, 1987; Bowker et al., 2010b; Petracci et al., 2012). Moreover, texture and appearance will be affected when over extracted meat is cooked, producing a ham-like appearance.

In general, injected whole birds, and injected then tumbled breast meat will produce better yields and texture than soaking alone. Brine formulation and ingredients used in the marinade play an important role in the effectiveness of the methods and yield retentions.

**Ingredients for Marinades**

Water, the main ingredient of marinades, and its quality are often times overlooked. Water that contains magnesium, calcium, and heavy metals is considered of poor quality, as it can interfere with functional ingredients such as phosphate. High water pH can cause precipitation of phosphate molecules, thus lowering the functionality of the phosphate (Oreskovich et al., 1992; Baluyot and Clark, 1996; Xiong and Kupski, 1999).

Phosphates, in particular sodium tri-polyphosphate (STPP), have been the functional ingredients of choice in marinades because of their ability to form chemical bonds by dissociating myofibrillar proteins and increasing meat pH and ionic strength, thus allowing the fixation of free water into bound water. Phosphates are extracted from
mineral rocks through acid extractions and are not considered an all-natural ingredient, but are generally recognized as safe (GRAS) by the Food and Drug Administration (FDA, 21 CFR 182.1781) (Baluyot and Clark, 1996; FSIS, 2005). Phosphates are controlled ingredients by the USDA and can be added to a maximum of 0.5 % of phosphate in the final food product (9 CFR 424.21). Performance of commercial phosphate blends is affected by the use of hard water, inadequate brine temperature, improper blend of phosphates (di-phosphates, tri-phosphates, STPP, hexametaphosphate, and pyrophosphate), and incorrect formulations (e.g. adding insufficient functional ingredients or using ingredients that counteract other ingredients) of the marinade (Baluyot and Clark, 1996; Alvarado and McKee, 2007).

Salts are also common functional ingredients in marinades. Salts (KCl and NaCl) are used to solubilize myofibrillar proteins, allowing more water to be bound to the muscle, thus improving sensorial attributes. The water binding ability of salts is similar to that of phosphates because it dissociates proteins due to its ionic strength, exposing more binding sites and increasing the meat’s WHC (Alvarado and McKee, 2007). Salts are not regulated, as their use is self-limiting to two percent, but higher percentages are reported to increase WHC in meats (Post and Heath, 1983; Alvarado and McKee, 2007; Saha et al., 2009). Health concerns such as hypertension also arise when higher amounts of salts are used to marinate poultry meats (Saha et al., 2009). Such concerns, together with cleaner label demands from consumers, have led to the development of niche markets for all-natural marinated meat products.
Research into the use of different blends of phosphates and salts has found that phosphates and salts have a synergistic effect: when used together, they increase the pick-up yield and produce lower drip loss and cook loss compared to when they are used individually.

**Natural Ingredients**

USDA’s Food Safety and Inspection services (FSIS) defines “all natural” label as an ingredient or product that has been minimally processed, does not contain artificial flavors or flavorings, color agents, chemical preservatives, or any other synthetic ingredient. The use of natural products in foods is a growing trend. This trend can be problematic for processors looking for commercially available all-natural ingredients that perform equally or better than phosphates. Phosphate replacers must be able to increase WHC, improve palatability, improve texture, and be labeled as an all-natural ingredient (Keeton, 1994; Alvarado and McKee, 2007; Barbut, 2007; Muhlisin et al., 2012).

Prior research has focused on products such as seaweed, tubers, barks, resins, cereals, and bacterial exudates, which can act as water binders in RTE and RTC products (Bater et al., 1993; Hachmeister and Herald, 1998; Barbut and Somboonpanyakul, 2007). They are commonly referred to as gums, fibers, and modified and unmodified starches. According to the FSIS definition of “all-Natural”, modified starches are not considered all-natural products as they have been treated with acids to modify their attributes. Some gums and fibers and non-modified starches are considered all natural.
and have been used as replacers of phosphate due to their ability to increase the water holding capacity, improve texture and yields, and reduce shrinkage (Keeton, 1994).

Barbut and Somboonpanyakul (2007) suggested that malva nut gum’s mode of action for binding water was through the retention of water in spaces within the protein gel network rather than water-protein interaction. When hydrated, starches create a network of spaces between the proteins where water is retained (Yackel and Cox, 1992; Bater et al., 1993; Hachmeister and Herald, 1998; Schilling et al., 2004; Barbut, 2009; Muhlisin et al., 2012). Hughes et al. (1997) reported that carrageenan gum and oat fiber increased WHC, reduced cook loss, and increased stability of batter in pork-beef frankfurter.

However, the use of these replacers is limited because they may mask desirable flavors, impart undesirable textures, and non-traditional ingredients are not usually accepted on labels by costumers (Keeton, 1994). Barbut and Somboonpanyakul (2007) reported that the use of malva nut gum at 0.6% lowers hardness of meat batters due to the high amount of gum hindering protein binding, thus reducing gel strength. Montero et al. (2000) found that xanthan gum decreases gel-forming ability of myofibrillar protein gels. Carrageenan gum has been reported to have a low melting point that may allow moisture loss over storage, and can also lower browning when cooked, affecting flavor development. Fibers, added in high quantities to formulation can impede meat particle agglomeration affecting texture, color, and flavor (Bater et al., 1993; Keeton, 1994; Hughes et al., 1997; Alvarado and Sams, 2004).
Sensory tests performed on phosphate replacers demonstrated consumer acceptability for these products when used in adequate amounts. Meat batters containing potato starch with 50% less fat were not significantly different from the control, as reported by Muhlisin et al. (2012). Bater et al. (1993) showed that carrageenan gum plus phosphate produced higher acceptability than control batter with just phosphates, but juiciness was higher in the phosphate control. Gums and fibers and non-modified starches may replace phosphates, provided they are used as recommended. New products are being created and tested, and are found to be as functional as phosphate blends.

SavorPhos is a proprietary blend of all natural flavorings, citrus flour, and less than 2% of sodium carbonate as a processing aid. It is currently being used as an all-natural ingredient for chicken marinades. Citrus flour is the functional fraction of the phosphate replacer that serves as a water binder. Citrus flour originates from various citrus pulps. The high fiber and low protein content are believed to contribute to the water holding capacity of the product (Fernandez-Gines et al., 2004; Aleson-Carbonell et al., 2005; Saricoban et al., 2008). The objective of this research project was to determine the effectiveness of SavorPhos as an all natural phosphate replacer in rotisserie whole birds and boneless/skinless breast fillets.
CHAPTER III
MATERIALS AND METHODS

Experiment One

A total of 100 Ross broilers were raised to 42 days of age at the Poultry Science Research, Teaching, and Extension Center (Texas A&M University, College Station). Birds were raised on a litter-lined floor and fed ad libitum starter, grower, and finisher corn/soybean meal base diet (NRC, 1994). Eight hours prior to slaughter, birds were removed from feed and allowed access to water ad libitum. Birds were then cooped and transported to the Poultry Processing Research Center and stunned at 13 to 15 mA for 5 to 7 s with an electrified knife (Cervin Electrical Systems, Minneapolis, MN) with an AC/DC converter set to 500Hz. After severing the left carotid artery and jugular vein the birds were bled for 90 s. Birds were then scalded for 45 s in 60°C water (Model SS-36-SS; Bower Corp., Haughton, IA), and feathers were picked in a rotary drum picker for 25 s (Model sp30ss; Bower Corp., Houghton, LA). Manual evisceration was performed prior to chilling (4°C) for 90 min in ice slush bath. Following chilling the RWOGs were stored in plastic tubs with lids inside a cooler at 4°C. The RWOGs were used at 24 h postmortem (PM). Raw weights and pH (Model IQ150; IQ Scientific Instruments, Inc., Carlsbad, CA; piercing probe PH77-SS.) of the individually tagged whole birds were obtained.
Four treatments were used in the study (Table 1) consisting of the two types of functional ingredients; the SavorPhos AF-200 and a commercially available phosphate blend (di- and tri-phosphates, hexameta phosphate), and two kinds of marinade blends, water and oil-water, formulated according to industry standards. For the two trials and two replications with in a trial a total of 50 birds were individually multi-needle injected (Inject-star BI-88 P-VSP; Mountain View, Arizona) at 20% w/w with each marinade type. The yield up-take (%) and pH were recorded immediately after injection, after 20 min drip time and at 24 h post-injection. Between sample times, the whole birds were stored in a cooler at 4°C. At 24 h post-injection, the marinated whole birds were netted and hung inside a smokehouse (Model MP-2; Koch equipment, Kansas City, MO). The whole birds were then cooked at 95°C with no smoke or steam until the internal temperature reached 73°C. Cooked carcasses were cooled at room temperature (90 min) to equilibrate temperatures before cook weights were measured to calculate cook loss. The amount of pick-up, retention, and cooking loss percentages were calculated.

**Table 1.** Marinade formulation percentages in whole birds and boneless/skinless breast fillets injected with either a phosphate blend or SavorPhos in water and oil-water marinades

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Water Marinade</th>
<th>Oil Marinade</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Phosphate Blend</td>
<td>Savorphos AF-200</td>
</tr>
<tr>
<td>Phosphate</td>
<td>0.40</td>
<td>0.00</td>
</tr>
<tr>
<td>Savorphos AF-200</td>
<td>0.00</td>
<td>0.50</td>
</tr>
<tr>
<td>Salt</td>
<td>0.70</td>
<td>0.70</td>
</tr>
<tr>
<td>Canola Oil</td>
<td>0.00</td>
<td>0.00</td>
</tr>
<tr>
<td>Meat</td>
<td>80.00</td>
<td>80.00</td>
</tr>
<tr>
<td>Total Batch</td>
<td>100.00</td>
<td>100.00</td>
</tr>
</tbody>
</table>
Experiment Two

One hundred and twenty-eight boneless/skinless breast fillets (~36 kg) were obtained from a local distributor 48 h PM and divided into 4 treatments (n=32/treatment) before being stored for 24 h in plastic tubes with lids in a cooler (4°C). Color measurements were obtained prior to injection from the bone-side of the fillets by averaging three readings using an L*a*b* scale of a calibrated colorimeter (Minolta Chroma Meter Model CR-200; Minolta Corp., Ramsey, NJ). Calibration was performed with the provided white tile. In addition, raw weights and pH (Model IQ150, IQ Scientific Instruments, Inc.; piercing probe PH77-SS.) were obtained from the individually tagged boneless/skinless breast prior to injection (Inject-star BI-88 P-VSP, Mountain View, AZ) with a 20% w/w solution (Table 1). Up-take percent was calculated after injection for the whole treatment. Per industry standard, marinade was added only to complete the 20% up-take target prior to vacuum tumbling for 20 min with 20 in of Hg at 25 rpm (Inject-star MC-25, Mountain View, AZ). The same marinade formulations were used from experiment one. They were comprised of two types of marinades: water and oil-water base, and within each, treatments SavorPhos and Phosphate blend were compared. Yield up-take percent and tumbled meat pH were recorded immediately after tumbling, 20 min after tumble, and again at 24 hr post-tumbling.

Color values for L*a*b* scale were collected using a three reading average of the bone-side of the fillets at 24 h post tumble (Minolta Chroma Meter Model CR-200, Minolta Corp.). Between sample times boneless/skinless breast were stored in cooler at
4°C. After data collection, BSB were baked at 177°C using a convection oven (Blodgett Zephaire G-1 speed; Blodgett Oven Co., Burlington, VT) as described in the method of Sams (1990). Individual BSB were put on metal racks in a 4-in high aluminum pan lined with foil. When boneless/skinless breasts internal temperature reached 73°C they were removed from the oven, weighed, individually wrapped with aluminum foil, and stored in the cooler overnight (4°C). Cooked weights were obtained after 24 h chilling to determine cook loss. Half of the cooked boneless/skinless breast were stored for texture and moisture analysis on the same day, while the other half were stored at 4°C for three days prior to sensory analysis. Formulas used to calculate injected pick-up, 20 min retention, 24 h retention, and cook loss/yield were the same as described previously.

Shear values (kg/g) were determined using the Instron Universal Testing Machine (Instron Corp., Canton, MA) using a 10 blade Allo-Kramer shear compression with a 500 kg load cell with a load range of 200 kg and a cross-head speed of 500mm/min (Sams, 1990). Moisture content was determined following AOAC methods 950.46 and 934.01,1998.

Sensory analysis was performed on half of the cooked BSB at the sensory testing facility at Texas A&M. A consumer taste panel was conducted with a triangle test designed to determine if consumers could detect differences between the treatments following the Meilgaard et al. (2007) procedure. Randomly selected panelists (n=50) were presented with two sets (water marinade and oil-water marinade) of samples displayed in a random fashion, two of SavorPhos and one phosphate blend while the
other half of the panelist were presented with two of phosphate blend and one of SavorPhos. Panelists were instructed to cleanse their palates in-between samples with distilled deionized water and unsalted saltine crackers. All samples were presented with a randomly generated 3-digit code in individualized booths that had controlled lighting and a hatched door through which the samples were served.

**Statistical Analysis**

Whole bird data were analyzed using the single-way ANOVA method from SigmaStat v3.1 (Systat Software 2003, San Jose, CA) and Tukey’s mean separation (P ≤ 0.05) to determine difference between treatments. Treatments within each of the marinades were compared but not between marinades or experiments. Boneless/skinless breast data were analyzed using the single-way ANOVA method using SigmaStat (Systat Software 2003) and Tukey’s mean separation (P ≤ 0.05) to determine difference between treatments. Treatments within each of the marinades were compared but not between marinades or experiments. Sensory results were considered to be different (P<0.05) if the numbers of correct responses from the panelist were higher than 22 out of 50 panelist (Meilgaard et al., 2007).
CHAPTER IV
RESULTS AND DISCUSSION

Experiment One

Water holding capacity is a desirable attribute for consumers and processors of rotisserie whole birds and is directly correlated with yield, and indirectly proportional to cook loss (Farr and May, 1970; Carpenter et al., 1979; Young and Lyon, 1997b, a; Alvarado and Sams, 2004; Bowker et al., 2010a). Retention of the injected brine impacts yield and quality traits of rotisserie birds. Thus, an increase in retained yield is favorable to the manufacturer and the consumer. Marination with multi-needle injectors is known to produce higher drip loss after the injection process (Owens, et al., 2009), possibly due to the openings created by the needles when the meat is penetrated. These openings may act as channels from which brine escapes.

Table 2 summarizes pick-up yields collected immediately after injection, 20 min retention, 24 h retention, and cooked loss percent in whole birds marinated with water and oil-water marinades. There was no significant difference between treatments in the pick-up %, at 20 min and 24 h retention %, and cook loss % in the water marinade treated whole birds. Therefore, SavorPhos AF200 performed equivalently to the phosphate blend when injected with water base marinades in whole birds.
Table 2. Pick-up %, 20 min retention %, 24 hr retention %, cook loss %, and pH values of rotisserie whole birds treated with either a phosphate blend or SavorPhos in water or oil-water marinades.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Yield</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Post-Inject</td>
<td>20 min Retention</td>
</tr>
<tr>
<td><strong>Water Marinade</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phosphate blend</td>
<td>23.24±5.01</td>
<td>97.68±1.09</td>
</tr>
<tr>
<td>Savorphos AF-200</td>
<td>25.08±2.85</td>
<td>98.05±0.61</td>
</tr>
<tr>
<td><strong>Oil Marinade</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phosphate blend</td>
<td>23.59a±4.67</td>
<td>98.13±0.86</td>
</tr>
<tr>
<td>Savorphos AF-200</td>
<td>26.26a±3.76</td>
<td>98.48±0.59</td>
</tr>
</tbody>
</table>

For each marinade, different letters within a column differ statistically (p≤0.05)
In the oil-water marinade, SavorPhos had higher pick-up and lower cook loss percent than the traditional phosphate blend, while retention 20 min and 24 hr post-injection was not different between the treatments. These results indicate that SavorPhos can replace the phosphate blend with improved pick up and reduced cook loss percentages. This gain in pick-up and reduced cook loss can be attributed to the citrus flour, a main functional component of SavorPhos. The citrus flour is a source of fiber and therefore can hold water and retain it during the cooking process (Keeton, 1994; Fernandez-Gines et al., 2004). The percent cook loss was significantly lower (p<0.05) and presented less variation than the oil-water phosphate blend marinated whole birds. Natural fibers, like the ones extracted from citrus albedo, have been used to increase the cooked yields. This increase is attributed to the ability of citrus albedo to bind oil and water in restructured and emulsion meat products (Iyengar et al., 1991; Jiminez-Colmenero, 1996; Mendoza et al., 1998b; Mendoza et al., 1998a; Fernandez-Gines et al., 2004). Lemon albedo used in conjunction with sodium tripolyphosphate and potato starch had better retention of moisture when compared to a control with no albedo in beef bologna (Fernandez-Gines et al., 2004). The similar performance attained by SavorPhos to the phosphate blend in water marinades, and the higher percent pick-up with lower cook loss percent of SavorPhos in oil-water marinades can be due to the citrus fiber as a functional ingredient.
The pH of the meat was measured as another WHC attribute of proteins. Table 2 indicates the results of the pH measurements. Research has indicated that chicken pH post mortem ranges between 5.7 and 6.0 (Owens et al., 2009; Gorsuch and Alvarado, 2010; Perlo et al., 2010; Petracci et al., 2012). As pH decreases closer to the isoelectric point (~5.3) of actin and myosin water binding ability is reduced, thus hindering the ability of meat to retain marinades post injection (Alvarado and McKee, 2007). For this experiment, there was no difference in the pH of the injected birds at raw, post-injection, 20 min, and 24 hr post injection in both treatments using water marinade (Table 2). However, in the oil marinade, the phosphate blends gave a higher pH at post injection compared to Savorphos AF-200. However, at 24 hr PM, there were no significant differences between the treatments so meat quality was not negatively affected as indicated by a better percent cook loss at 24 hr PM. Prior research on salt and phosphate marinated breast fillets shows an increase in pH after marination (Gorsuch and Alvarado, 2010; Perlo et al., 2010). The use of high pH (>9.0) phosphate marinades demonstrated an increase in broiler pectoralis pH (Alvarado and Sams, 2004). Sheard and Tali (2004) reported an increase of 0.5 in pH on injected pork loins compared to loins injected just with salt. Saricoban et al. (2008) reported no change in pH in mechanically deboned chicken meat batters containing lemon albedo (2.5%), salt (2.5%), and phosphate (0.25%) when compared to a control containing no albedo.
Overall SavorPhos performed equally or better than a phosphate blend on yield results, when used in a water or oil-water marinade for injection rotisserie whole birds. Therefore, Savorphos can be used as a replacement for phosphates when consumers are concerned about clean labels.

Experiment Two

Up-take and retention yield positively impact consumers quality perception as well as manufactures margins (Hargett et al., 1980; Heath and Owens, 1987; Owens et al., 2009; Gorsuch and Alvarado, 2010; Perlo et al., 2010; Petracci et al., 2012). Boneless/skinless breast fillets yield parameters (Table 3) were measured at post-marination, 20 min and 24 hr post-marination, and after cooking. In water-based marinades, the boneless/skinless breast treatments were similar in percent pick-up, 20 min and 24 hr retention. Additionally, percent cook loss was decreased for SavorPhos when compared to the phosphate blend. In the oil-water marinades, percent pick-up was significantly higher for SavorPhos when compared to the phosphate blend, but showed no difference in 20 min, 24 h percent retention and cook loss. These results indicate that
in oil-water marinades SavorPhos performs similarly to the phosphate blend in retention
% and cook loss %. This observation agrees with the finding described by Allen et al.
(1998) and Bowker et al. (2010) who reported yield loss from tumble marinated breast
fillets stored between 1 and 3 days. The use of citrus fiber (citrus albedo) is well
documented in other types of meat products, and shows similar results as the ones
observed in this study. Aleson-Carbonell et al. (2005) found that beef patties formulated
with dry cooked lemon albedo and salt had higher cook yields (73.27%) than patties
without the lemon albedo (62.31%). Results described by Aleson-Carbonell et al. (2005)
suggested that lemon albedo fiber added to breakfast fresh sausage achieved a higher
moisture retention, reduced cooking loss, and retained more fat when compared to a
sausage that contained no albedo. Marin et al. (2003) reported that the insoluble fiber of
citrus albedo is responsible for the lipid holding capacity, while the soluble portion is
responsible for the WHC of the product.
Table 3. Pick-up %, 20 min retention %, 24 hr retention %, cook loss %, and pH values of bonless/skinless breast fillets treated with either a phosphate blend or SavorPhos in water or oil-water marinades.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Yield</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Post-Inject</td>
<td>20 min Retention</td>
</tr>
<tr>
<td>Water Marinade</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phosphate blend</td>
<td>19.11±2.26</td>
<td>99.41±0.63</td>
</tr>
<tr>
<td>Savorphos AF-200</td>
<td>19.45±3.21</td>
<td>99.34±0.43</td>
</tr>
<tr>
<td>Oil-water Marinade</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phosphate blend</td>
<td>19.11&lt;sup&gt;b&lt;/sup&gt;±2.27</td>
<td>99.41±0.63</td>
</tr>
<tr>
<td>Savorphos AF-200</td>
<td>24.09&lt;sup&gt;a&lt;/sup&gt;±2.81</td>
<td>99.37±0.33</td>
</tr>
</tbody>
</table>

*<sup>a</sup>-<sup>b</sup> For each marinade, different letters within a column differ statistically (p≤0.05)
The pre and post-marination pH of the meat can impact the water holding capacity (Alvarado and McKee, 2007). The lower the meat pH pre-marination, the more predisposed the meat is to retain less water due to low isoelectric charges within the myofibrillar protein. The pH of the boneless/skinless breast fillets (Table 3) marinated (water) with a phosphate blend was not different when compared to the SavorPhos for the raw meat. The water marinade pH post-injection, 20 min, and 24 hr retention were higher on all sample times for the phosphate blend compared to SavorPhos. In the oil-water marinades, the same differences were observed. Even though the pH results are lower than the phosphates in both treatments, the pH values are still within normal meat range for good meat quality (Alvarado and Sams, 2004; Alvarado and McKee, 2007). These results are in agreement with prior research where the phosphate blend increased the pH of the meat after marination and lemon albedo maintained the meat batter pH on dry sausage (Sheard and Tali, 2004; Alvarado and McKee, 2007; Saricoban et al., 2008; Perlo et al., 2010). Phosphates increase meat pH (Alvarado and McKee, 2007), even though SavorPhos decreased meat pH post –injection, this decrease in pH did not negatively impact meat quality (Table 3).

Meat pH values have been correlated to color measurements (L*- lightness value) of boneless/skinless breast, where the lighter the color the lower the pH (Alvarado and McKee, 2007; Barbut, 2009; Owens et al., 2009). Color variation of meat is of great importance to the consumer acceptability and meat functionality (Alvarado and McKee, 2007; Barbut, 2009; Owens et al., 2009; Gorsuch and Alvarado, 2010). Barbut (2009) reported that consumers become concerned when boneless/skinless breast meat is too
pale or too dark. Wilkins et al. (2000) stated that color extremes are likely to be
discriminated at the point of purchase. Color is measured in the L*a*b* scale where L*
represents the lightness, a* the redness, and b* the yellowness of meat. As expected, raw
meat color did not differ in L*, a*, and b* values in both the water and oil-water
marinades between the two treatments (Table 4). At 24 hr post injection the oil-water
marinade treatments were also not statistically different in L*, a* and b*. However, at 24
hr post-injection in the water marinade, the phosphate blend a* values were higher when
compared to the SavorPhos indicating a more reddish color. Also, the b* value were
significantly higher in the SavorPhos treatment when compared to the phosphate
treatment indicating a more yellow appearance. However, these objective differences
were not observed in the consumer panel and these differences may not be discernible by
consumers. Prior research has shown an increase in lightness values 3 hr after
marination with a phosphate blend possibly due to the increase in extracellular water
(Gorsuch and Alvarado, 2010). This L* value increase was not observed in the current
experiment. Fernandez-Lopez et al. (2004) reported an increase of all color values in
bologna made with citrus albedo. The yellowness in the citrus fiber may explain the
increase of the b* value of 24 hr water marinated fillets. Aleson-Carbonell et al. (2005)
reported similar finding in the internal color of cooked beef patties. Though differences
for a* and b* values (P<0.05) were found for water marinated breast fillets 24 hr post
injection boneless/skinless breast fillets, industry application would not be impacted as
one of the main drivers of costumer acceptance of raw breast meat is the lightness
Moisture content on cooked boneless/skinless breast was measured as a WHC parameter (Table 4). No difference (P<0.05) was found between SavorPhos and the phosphate blend for both types of marinades (water and oil-water). These results are interesting as SavorPhos had a decreased percent cook loss in the water marinade. Lee et al. (2008) reported similar results where two natural marinated chicken breasts, one with carrageenan and the other without, showed the same moisture content (71.9 and 70.8, respectively) between each other but cook loss (19.0% vs 24.1%) was lower for the marinade containing carrageenan. These results indicate that SavorPhos can replace phosphate without negatively affecting total moisture.

Texture was analyzed to determine tenderness of the marinated boneless/skinless breast fillet. Prior research has correlated texture values with consumer perception of tenderness (Saha et al., 2009). SavorPhos-treated meat had significantly lower texture values, indicating more tender meat when compared to meat treated with the phosphate blend in both the water marinade and the oil-water marinade. This increased tenderness can be explained by the lower cook loss for water marinade treatments and the possible swelling effect of the citrus flour. Another explanation could be related to the swelling of the citrus fiber within the meat marinated with SavorPhos, which might cause an increase in tenderness (Hughes et al., 1997).
Table 4. Boneless/skinless breast fillets Moisture %, Texture, and Color of raw meat and 24hr post marination, treated with a water and water-oil marinade.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Color - Raw</th>
<th></th>
<th></th>
<th>Color – 24hr</th>
<th></th>
<th></th>
<th>Moisture (%)</th>
<th>Texture (Kg/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>L*</td>
<td>a*</td>
<td>b*</td>
<td>L*</td>
<td>a*</td>
<td>b*</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Water Marinade</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phosphate Blend</td>
<td>55.10</td>
<td>1.67</td>
<td>5.86</td>
<td>52.00</td>
<td>2.33&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.16&lt;sup&gt;b&lt;/sup&gt;</td>
<td>71.49±1.91</td>
<td>6.75&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Savorphos AF-200</td>
<td>57.54</td>
<td>1.36</td>
<td>7.96</td>
<td>54.68</td>
<td>1.25&lt;sup&gt;b&lt;/sup&gt;</td>
<td>6.82&lt;sup&gt;a&lt;/sup&gt;</td>
<td>72.23±2.25</td>
<td>5.92&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Oil-water Marinade</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phosphate Blend</td>
<td>58.62</td>
<td>2.09</td>
<td>6.99</td>
<td>55.32</td>
<td>1.12</td>
<td>3.95</td>
<td>70.20±1.67</td>
<td>6.14&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Savorphos AF-200</td>
<td>57.21</td>
<td>2.40</td>
<td>5.80</td>
<td>54.68</td>
<td>1.41</td>
<td>4.96</td>
<td>69.74±1.61</td>
<td>4.94&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a-b</sup> For each marinade, different letters within a column differ statistically (p≤0.05).
A consumer (n=50) triangle test was performed to establish if consumers could distinguish between the SavorPhos and the phosphate blend in water and oil-water marinades of cooked boneless/skinless breast fillets. The consumer panel was not able to determine significant differences (P<0.05) between the treatments (SavorPhos and Phosphate blend) with in each of the marinades. Difference was establish when 22 or more out of the 50 panelist were able to determine the similar sample on the triangle test, with an α=0.05. Therefore, less than 22 panelists were able to distinguish between the two similar samples for each of the two marinades indicating SavorPhos can be used as a natural replacement for phosphates without negatively affecting consumer preferences.
The use of SavorPhos in water and oil-water base marinades will allow an equally or enhanced performance of pick-up and retention yields, as well as cook loss % when compared to a phosphate blend in rotisserie chickens and boneless/skinless breast meat. Allowing industry to effectively use SavorPhos Af-200 as an all-natural replacer of commercial di- and tri-phosphates, and hemametaphosphate blends. Lower or similar variations were observed on the rotisserie whole birds and bonellee/skinless breast fillets yield percentages for SavorPhos treatments in both water and oil-water marinades. Texture values of boneless/skinless breast meat are improved with the use of SavorPhos and without negatively affecting color or consumer preferences. This makes SavorPhos a potential all-natural replacer of phosphate blends for use in boneless/skinless breast meat, as well as rotisserie whole birds.

The ability of SavoPhos to act as a replacer of phosphates blends together with the enhanced texture and lower variability, allows for a more consistent and desirable product in the processing environment. As variation in uptake yields is reduced cooking, chilling, and freezing become more consistent achieving a higher quality and safer product for either ready-to-cook or ready-to-eat poultry meat. Offering a cleaner all-natural label rotisserie whole bird or RTC and RTE chicken breast fillet to the consumer.
REFERENCES


NCC 2012. How broilers are marketed.


