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Effect of NaCl Concentration and Cooking Temperature on the Color and Pigment Characteristics of Presalted Ground Chicken Breasts

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Abstract This study was conducted to determine the effects of NaCl concentration and cooking temperature on the color and pigment characteristics of presalted ground chicken breasts. Four treatments with different salt concentrations (0%, 1%, 2%, and 3%) were prepared and stored for 7 d prior to cooking. Each sample was cooked to four endpoint temperatures (70°C, 75°C, 80°C, and 85°C). The salt concentration affected the color and pigment properties of the cooked ground chicken breasts. As the salt concentration increased, the cooking yield and residual nitrite content also increased. However, the samples with 1%, 2%, and 3% NaCl showed similar nitrosyl hemochrome and total pigment contents. Among the products containing salt, the samples with 3% NaCl showed the lowest percentage myoglobin denaturation (PMD) and the lowest CIE a* values. The cooking temperature had limited effects on the pigment properties of cooked ground chicken breasts. The oxidation-reduction potential and residual nitrite contents increased with cooking temperature, while the PMD, nitrosyl hemochrome, total pigment contents and CIE a* values were similar in the samples cooked at different temperatures. These results indicated that the addition of up to 2% salt to ground chicken breasts and storage for 7 d could cause the pink color defect of cooked products. However, the addition of 3% NaCl could reduce the redness of the cooked products.

Keywords ground chicken breast, NaCl, cooking temperature, pink color, pigment properties

Introduction

Salt (NaCl) is one of the oldest food ingredients and the most important and fundamental one in various meat products. The major functions of NaCl are flavor enhancement, shelf-life extension through microbial growth control, increasing water holding capacity, and texture improvement (Sebranek and Fox, 1991; Sebranek, 2009). Further, the addition of NaCl to meat products has been shown to affect meat color. NaCl may decrease the heat stability of myoglobin and hemoglobin (Ahn and Maurer,

1989c), and excessive addition of NaCl could accelerate the oxidation of myoglobin, thus reducing the redness of the cooked meat (Min et al., 2009; Jeong, 2017; Trout, 1989). In cured meat, chloride ions dissociated from NaCl are known to increase the conversion of nitrite to nitric oxide, thus accelerating the formation of cured meat color (Sebranek and Fox, 1991).

The final cooking temperature is also an important factor that influences the color of cooked meat. Generally, the degree of redness of the cooked product decreases when the final cooking temperature is increased (Girard et al., 1989; Helmke and Froning, 1971; Howe et al., 1982). This reduction in redness is caused by the denaturation of myoglobin and hemoglobin during cooking (Girard et al., 1989). For example, as the final cooking temperature for turkey breast meat increases, myoglobin denaturation increases, and consequently, redness is reduced (Girard et al., 1989; Davis and Franks, 1995; Trout, 1989). Trout (1989) demonstrated that, when turkey breast was cooked to temperatures less than 76°C, it resulted in incomplete denaturation of myoglobin, while cooking temperatures more than 76°C caused the formation of a pink hemochrome. However, Girard et al. (1990) reported that both turkey breast and pork loin, when cooked to higher than 85°C, showed the pink pigment, suggesting that reduced cytochrome c may be responsible for the pink color. The presence of NaCl increases the stability of this pigment at 80°C or higher and this may play a very important role in the development of pink color in cooked meat (Ahn and Maurer, 1989c).

Pink color defect is a color quality problem in cooked meat that has been reported in a wide variety of poultry products. This problem occurs sporadic and factors affecting this phenomenon are complex and varied. Consumers would think that the product is still under-cooked when appearing pink color present in fully cooked meat products, although this phenomenon is not a food safety issue (Friesen and March, 2000; Suman and Joseph, 2014). Several studies have been conducted to solve and control this problem, and undenatured myoglobin, oxymyoglobin, nitrosyl hemochrome, carbon monoxide, and cytochrome c have been reported to cause the pink color (Ahn and Maurer, 1989a; Ahn and Maurer, 1989c; Bernofsky et al., 1959; Davis and Franks, 1995; Ghorpade and Cornforth, 1993; Girard et al., 1990; Heaton et al., 2000; Ledward, 1971; Nam and Ahn, 2002; Trout, 1989). In addition, it is also possible that the pink color is caused by handling and processing conditions of the carcass, such as feed, transport condition, slaughtering gas environment, cooking temperature, cooling rate, storage period, and added materials (Claus et al., 1994; Froning et al., 1969a; Froning et al., 1969b; Sackett et al., 1986). Previous studies have mostly attempted to create the pink color by adding pink-generating ligands such as nitrite and nicotinamide or by simulating the reducing ability of pink color (Ahn and Maurer, 1989a; Ahn and Maurer, 1990a; Claus et al., 1994; Sammel and Claus, 2003; Sammel and Claus, 2006; Sammel and Claus, 2007; Schwarz et al., 1997; Schwarz et al., 1999; Slesinski et al., 2000a; Slesinski et al., 2000b). Recently, Claus and Jeong (2018) reported that an intense pink color was naturally produced in fully cooked ground turkey breasts by certain processing procedures (presalting with 2% NaCl and storage for 7 d before being cooked) without the addition of any pink-generating ligands. They suggested that a synergistic effect between the storage of ground turkey and the presence of salt promoted the pink color associated with the formation of reduced nicotinamide-denatured globin hemochrome. These findings could provide the means for controlling the pink color and understanding the mechanism behind color development in meat products. In their study, however, the effects of the salt addition level coupled with cooking temperature under the processing condition that promote intensive pink color in cooked turkey breasts have been not established.

Therefore, this study was conducted to investigate the effects of salt concentration and endpoint cooking temperature on color and pigment characteristics of ground chicken breasts as baseline data for the development of pink color in meat products.

Materials and Methods

Processing and preparation

The processing was performed with slightly modified procedure from a previous study (Claus and Jeong, 2018). Fresh, skinless, boneless chicken breasts (1 d postmortem) were obtained from a local processor (Kwangsung Food, Korea). Raw material was shipped in an insulated cooler and refrigerated (2–3°C) until use. Three separate replicates of ground chicken breast were used in this study. From each replication, a representative sample of raw ground chicken was used for pH determination and residual nitrite content analysis. A total of 20 kg of raw chicken trimmings was used for each replication and ground with a 0.3 cm plate using a chopper (TC-22 elegant plus, Tre Spade, Italy). The ground meat was randomly divided into four batches (4 kg each), based on NaCl concentrations (0%, 1%, 2%, and 3% NaCl). Each batch of ground meat was mixed with 0%, 1%, 2%, or 3% NaCl using a mixer (5K5SS, Whirlpool Inc., USA) for 10 min and individually vacuum-packaged with a vacuum packaging machine (M6-TM, Leepack Co., Ltd., Korea) in polyethylene/nylon bags, which were then stored for 7 d under refrigeration (2–3°C). After 7 d, each batch was remixed (5K5SS, Whirlpool Inc., USA) for 5 min, and air pockets were removed in a vacuum chamber (M6-TM, Leepack Co., Ltd., Korea); the batches were then stuffed into conical centrifuge tubes (approximately 50 g each). The tubes were centrifuged at 2,000×g for 10 min (FELTA5, Hanil Science Corp., Korea) to remove air pockets. All the samples were then cooked to four different endpoint temperatures (70°C, 75°C, 80°C, and 85°C, respectively) in a 90°C water bath (CB60L, Dongwon Scientific Machinery Corp., Korea) and the temperature was monitored by randomly placing four thermocouples attached to a 4-channel digital thermometer (Tes-1384, Ketech Scientific Instrument Co., Ltd., Taiwan) in the center of the extra samples in the water bath. After cooking, the samples were immediately cooled on ice for 20 min and stored at 2–3°C overnight in the dark, until further analysis. The experiments were replicated thrice.

Cooking yield, pH, and oxidation-reduction potential (ORP) determination

Stuffed ground chicken meat samples were weighed prior to cooking to determine the raw sample weight. Cooked weights were measured prior to color measurements to determine yield. Cooking yield was calculated as: $[\text{cooked sample weight}/\text{raw sample weight}] \times 100$. A pH meter (Accumet AB50, Thermo Fisher Scientific Inc., Singapore, Singapore) was used to measure the pH of 5 g of raw meat or cooked chicken samples homogenized in 25 mL of distilled, deionized water. ORP of cooked turkey products was measured, following the modification of Cornforth et al. (1986) and John et al. (2005). A sample (10 g) from each chicken product was homogenized (DI 25 basic, IKA-Werke GmbH & Co. KG, Germany) with 20 mL of 0.1 M sodium carbonate for 15 s at 13,000 rpm. Fifty microliters of butylated hydroxytoluene (BHT, 7.2% in ethanol) was added to minimize sample oxidation during blending. The ORP values were determined after 3 min of stabilization using a platinum Ag/AgCl combination electrode (Model 13-620-631, Thermo Fisher Scientific Inc., Singapore) attached to the pH meter set to the milli-volt scale.

Instrumental color determination

CIE (L*, a*, b*) values were measured on freshly cut surfaces of cooked samples using a chroma meter (CR-400, 8 mm aperture, illuminant C; Konica Minolta Corp., Japan) calibrated with a white plate (L* 94.90, a* -0.39, b* 3.88). CIE (Commission Internationale de l'Eclairage) measurements were used to determine lightness (CIE L*), redness (CIE a*), and yellowness (CIE b*). Two slices were cut parallel to the longitudinal axis of each ground chicken breast sample, and 3

measurements per slice (six readings per sample) were taken immediately after cutting.

Myoglobin content, percentage myoglobin denaturation (PMD), and pigments determination

Myoglobin (Mb) was extracted from uncooked or cooked turkey breast products using the method published by Warriss (1979) and Trout (1989). The extracted supernatants were clarified by filtration through Whatman No. 1 filter paper, and absorbance was determined at 525, 572, and 700 nm (Krzywicki, 1979) using a UV/VIS spectrophotometer (UV-1800, Shimadzu Corp., Kyoto, Japan). The total myoglobin (Mb) and PMD were calculated using the following formulas (Trout, 1989): $Mb \text{ (mg/g)} = (A_{525} - A_{700}) \times 2.303 \times \text{dilution factor}$, $PMD = [1 - (Mb \text{ concentration after heating} / Mb \text{ concentration before heating})] \times 100$. To obtain the percentage reflectance, a UV/VIS spectrophotometer (UV-1800, Shimadzu Corp., Japan) was used to read absorbance on the filtrate from 400 to 700 nm with 1-nm increments. Four absorbance readings were taken on each cooked ground chicken sample, and the absorbance data were converted to percentage reflectance using the equation described by Stewart et al. (1965). Nicotinamide hemochrome (rNIC) was estimated by the percent reflectance ratio of $\%R_{537} \text{ nm} / \%R_{553} \text{ nm}$, where a higher value indicates more pigments (Schwarz et al., 1998).

Nitrite, nitrosyl hemochrome, and total pigment analysis

Representative fresh ground turkey samples from each replication and cooked products from each NaCl treatment with the different endpoint cooking temperatures were analyzed for residual nitrite by the method described by AOAC (2007). Nitrosyl hemochrome and total pigments were measured on cooked turkey samples after extraction in 80% acetone and acidified acetone (Hornsey, 1956). For nitrosyl hemochrome determination, 10 g of cooked turkey product was blended with 40 mL acetone and 3 mL distilled, deionized water using a homogenizer (Polytron PT10-35, Kinematica AG, Switzerland). The homogenized samples were kept in the dark for 15 min before absorbance measurement. The homogenate was filtered through a Whatman No. 1 filter paper, and absorbance of the filtrate at 540 nm (A_{540}) was determined using a spectrophotometer (Model UV mini 1240, Shimadzu Corporation, Japan). Nitrosyl hemochrome concentration (ppm) was calculated as: $A_{540} \times 290$. For total pigment measurement, 10 g of the cooked samples was blended with 40 mL acetone, 1 mL HCl, and 2 mL distilled, deionized water, kept in the dark at cold temperatures (2–3°C) for 1 h, and then filtered through a Whatman No. 1 filter paper. Absorbance was measured at 640 nm (A_{640}). The total pigment concentration (ppm) was calculated as: $A_{640} \times 680$.

Statistical analysis

The experimental design of this study was a split plot design, with the four NaCl concentrations (0%, 1%, 2%, and 3%) representing the whole plot factor and the four endpoint cooking temperatures (70°C, 75°C, 80°C, and 85°C) representing the split plot factor. The fixed effects for NaCl concentration and endpoint temperature and their interactions were performed using the PROC MIXED procedure of the SAS 9.4 software (SAS, 2012). Dependent variable means were separated ($p < 0.05$) by pairwise comparisons using the pdiff option. The experiment was replicated thrice.

Results and Discussion

The significance of two effects, NaCl concentration (N) and cooking temperature (C), and their interactions on color and pigment characteristics is presented in Table 1. Two-way interactions between the main effects (N×C) were found for cooking

Table 1. Significance of main and interaction effects of NaCl concentrations and cooking temperature on color and pigments in cooked ground chicken breasts

Main effects and interaction	Dependent variables											
	Cooking yield	pH	ORP	Nitrite	Myoglobin	PMD	Nitrosyl hemo-chrome	Total pigments	CIE Color			rNIC
									L*	a*	b*	
NaCl concentration ¹ , N	**	**	**	**	**	**	**	**	**	**	**	**
Cooking temperature ² , T	**	NS	*	*	NS	NS	NS	NS	**	NS	**	NS
N×T	**	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS

¹ All raw ground meats were salted (0, 1, 2, and 3% NaCl, respectively) and stored for 7 d before being cooked.

² The samples were cooked to an internal temperature of 70, 75, 80, and 85°C, respectively, in a 90°C water bath.

* $p < 0.05$; ** $p < 0.0001$; NS, not significant.

ORP, oxidation reduction potential; Nitrite, residual nitrite contents; Myoglobin, amount of undenatured myoglobin; PMD, percentage myoglobin denaturation; CIE L*, lightness; CIE a*, redness; CIE b*, yellowness; rNIC, reflectance estimator of nicotinamide hemo-chrome (%R537 nm/%R553 nm).

yield ($p < 0.0001$) and total pigment contents ($p < 0.05$).

pH values and residual nitrite content for raw ground chicken breast

The average pH value of the raw ground chicken breast samples was 5.96 (SD=0.12). A representative sample of raw ground chicken for all three replicates contained 0.16 ppm residual nitrite (SD=0.04). These results were consistent with those of Jeong (2017), who reported that raw ground chicken breast had an average pH of 5.98 and 0.18 ppm of residual nitrite. Claus and Jeong (2018) also reported similar results for raw turkey breast meat.

Effect of NaCl concentration

NaCl concentration affected all dependent variables tested in this study, associated with color and pigment properties of cooked ground chicken breasts (Table 1).

The cooking yield was lowest in 0% NaCl treatment ($p < 0.05$), and increased ($p < 0.05$) as the NaCl concentration increased from 0% to 3% (Table 2). This is consistent with the results of Lopez et al. (2012), who found that cooking loss decreased as NaCl concentration increased in vacuum-tumbled broiler breast meat. These results suggested that the addition of NaCl increased the negative charge on the protein surface, which increases the water holding capacity (Sebranek, 2009).

The pH values of cooked ground chicken products decreased with NaCl concentration ($p < 0.05$), but there were no significant differences ($p > 0.05$) in pH values between products treated with 2% and 3% NaCl (Table 2). This result was similar to that of Puolanne et al. (2001), who found that the addition of NaCl reduced the pH of pork and beef sausages. Our results on the effect of salt on the pH of cooked chicken breast meat are also partially consistent with those of Jeong (2017), who reported that the pH of cooked chicken breast meat was not affected by varying the salt level from 0 to 3%.

The ORP of meat affects its color through the formation of hemochromes, which is promoted by reducing conditions (Cornforth et al., 1986) and strong reducing conditions are also essential for the formation of a stable pink color in cooked meats (Cornforth et al., 1991). Therefore, measuring ORP is important for investigating the color characteristics. In this study, ORP was found to be lowest ($p < 0.05$; most reducing condition) in cooked ground chicken products without salt (0% NaCl).

Table 2. Effects of NaCl concentration on cooking yield, pH, oxidation reduction potential (ORP) and residual nitrite contents in cooked ground chicken breasts

NaCl concentrations ¹	Dependent variables			
	Cooking yield (%)	pH	ORP (mV)	Nitrite (ppm)
0%	89.5 ^C	6.19 ^A	-109.9 ^C	0.27 ^D
1%	97.0 ^B	6.16 ^B	-106.6 ^B	0.39 ^C
2%	97.4 ^{AB}	6.13 ^C	-103.6 ^A	0.51 ^B
3%	97.9 ^A	6.13 ^C	-104.8 ^A	0.56 ^A
(SE)	(0.55)	(0.09)	(5.92)	(0.05)

¹ All raw ground meats were salted (0, 1, 2, and 3% NaCl, respectively) and stored for 7 d before being cooked.

^{A-D} Means within a column with different superscript letters are significantly different ($p < 0.05$).

ORP, oxidation reduction potential; Nitrite, residual nitrite contents; SE, standard error.

As the concentration of NaCl increased in the ground chicken breast, the ORP also increased ($p < 0.05$) (Table 2). This result was in contrast with that of Ahn and Maurer (1989b), who reported that ORP decreased upon the addition of salt in ground turkey meat. This disagreement may be related to differences in the pH values in the two studies. Higher pH has been shown to promote reducing conditions (Antonini and Brunori, 1971; Claus and Jeong, 2018). Samples without NaCl had the highest pH values, resulting in the most negative ORP values. In addition, there were no significant differences in the pH of samples treated with 2% and 3% NaCl (Table 2). Jeong (2017) found that cooked ground chicken breasts with 0, 1, 2, or 3% NaCl had similar ORP values, when they were stored for 3 d before cooking, although these values tended to slightly decrease with increasing NaCl concentrations.

Residual nitrite contents in cooked ground chicken breasts were found to increase ($p < 0.05$) with increasing NaCl concentrations (Table 2). It seems likely that NaCl caused the increase of residual nitrite (Ahn and Maurer, 1989a). However, the detected levels of residual nitrite contents ranged from 0.26 to 0.56 ppm across all samples in this study. These values were not sufficient to develop a pink color in meat products, as reported by previous researches (Ahn and Maurer, 1989a; Heaton et al., 2000). Ahn and Maurer (1989a) found that addition of 1 ppm nitrite resulted in a significant increase in the redness of turkey breast meat. Heaton et al. (2000) also reported that the minimum amount of nitrite for detecting pinkness was 1.3 and 0.7 ppm for cooked turkey and chicken breast, respectively.

The myoglobin contents were reduced ($p < 0.05$) by the addition of NaCl, with the exception of 3% NaCl. However, there was no significant difference ($p > 0.05$) between the myoglobin contents in the samples treated with 1% and 2% NaCl (Table 3). This finding is consistent with that of Jeong (2017), who also reported a decrease in myoglobin content when salt concentration was increased from 0% to 2%, except for cooked chicken breasts with 3% NaCl. However, we found no significant differences in the myoglobin contents of samples treated with 1% and 2% NaCl ($p > 0.05$). Samples treated with 3% NaCl had lower ($p < 0.05$) myoglobin contents than those treated with 1% and 2% NaCl, but had similar myoglobin content ($p > 0.05$) as the samples without NaCl. The effects of salt on the PMD contrasted with those on myoglobin content (Table 3). The addition of NaCl is known to destabilize myoglobin in meat products upon cooking, leading to greater heat denaturation (Ahn and Maurer, 1989c; Jeong, 2017; Min et al., 2009; Trout, 1989). Claus and Jeong (2018) reported that cooked ground turkey breasts with 2% NaCl had lower myoglobin contents and higher PMD than those without NaCl, regardless of the processing condition, which is similar to our results.

Both nitrosyl hemochrome and total pigment content was highest in samples without NaCl ($p < 0.05$), although no

Table 3. Effects of NaCl concentration on myoglobin contents, percentage myoglobin denaturation (PMD), nitrosyl hemochrome and total pigments in cooked ground chicken breasts

NaCl concentrations ¹	Dependent variables			
	Myoglobin (cooked, mg/g)	PMD (%)	Nitrosyl hemochrome (ppm)	Total pigments (ppm)
0%	0.24 ^A	79.53 ^B	2.40 ^A	15.53 ^A
1%	0.20 ^B	84.17 ^A	1.38 ^B	14.46 ^B
2%	0.20 ^B	84.84 ^A	1.46 ^B	14.61 ^B
3%	0.24 ^A	79.02 ^B	1.12 ^B	14.38 ^B
(SE)	(0.01)	(0.77)	(0.29)	(0.29)

¹ All raw ground meats were salted (0, 1, 2, and 3% NaCl, respectively) and stored for 7 d before being cooked.

^{A,B} Means within a column with different superscript letters are significantly different ($p < 0.05$).

Myoglobin, amount of undenatured myoglobin; PMD, percentage myoglobin denaturation; SE, standard error.

significant difference ($p > 0.05$) was observed between the samples treated with NaCl (Table 3). Ahn and Maurer (1989a) reported that the addition of NaCl reduced the nitrosyl hemochrome and total pigment in turkey meat treated with nitrite, which is consistent with our results. Jeong (2017) also obtained similar results for cooked ground chicken breasts when ground meat was presalted (0-3% salt) and stored for 3 d before cooking.

The CIE L* values decreased with increasing NaCl concentration ($p < 0.05$). The CIE a* value was lower in samples treated with 3% NaCl ($p < 0.05$) than in the others. However, samples treated with 0%, 1%, and 2% NaCl had similar ($p > 0.05$) CIE a* values in cooked chicken breasts (Table 4). Holownia et al. (2003) reported that pink color can be detected when a* value is more than 3.8 in chicken breast meat. In this study, CIE a* values of samples treated with 1% and 2% NaCl were 4.08 and 3.83, respectively. Therefore, the pink color was developed without any pink-generating ligands in cooked ground chicken breasts. In addition, Claus and Jeong (2018) demonstrated that the storage of ground turkey in the presence of 2% NaCl, rather than without salt, promoted the formation of pink pigments. Although the absence of salt in ground chicken meat was tested in this study, the average CIE a* value of these samples was 3.67, which was similar to those of the samples treated with 1% and 2% NaCl (Table 4). The CIE b* values were highest ($p < 0.05$) in the samples without NaCl, and tended to

Table 4. Effects of NaCl concentration on CIE L*, CIE a*, CIE b*, and rNIC in cooked ground chicken breasts

NaCl concentrations ¹	Dependent variables			
	CIE L*	CIE a*	CIE b*	rNIC
0%	80.99 ^A	3.67 ^A	9.04 ^A	1.007 ^B
1%	78.19 ^B	4.02 ^A	8.20 ^C	1.009 ^A
2%	76.44 ^C	3.83 ^A	8.18 ^C	1.008 ^B
3%	75.35 ^D	2.60 ^B	8.70 ^B	1.003 ^C
(SE)	(0.41)	(0.33)	(0.35)	(0.0013)

¹ All raw ground meats were salted (0, 1, 2, and 3% NaCl, respectively) and stored for 7 d before being cooked.

^{A-D} Means within a column with different superscript letters are significantly different ($p < 0.05$).

CIE L*, lightness; CIE a*, redness; CIE b*, yellowness; rNIC, reflectance estimator of nicotinamide hemochrome (%R537 nm/%R553 nm); SE, standard error.

decrease ($p < 0.05$) with increasing NaCl concentration (Table 4). No significant differences ($p > 0.05$) were observed in the CIE b^* values of samples treated with 1% and 2% NaCl. The samples treated with 3% NaCl showed higher ($p < 0.05$) CIE b^* values than those treated with 1% and 2% NaCl. Ahn and Maurer (1989a) reported that, when 2% NaCl was added to turkey breast, the L^* value decreased and a^* value increased, which is consistent with our results. The addition of more than 3% NaCl may reduce the redness of cooked products by promoting oxidative conditions and denaturing myoglobin (Jeong, 2017; Rhee and Ziprin, 2001; Trout, 1989), and this could explain the decrease in the CIE a^* value of samples treated with 3% NaCl in this study.

Nicotinamide can form strong heme complexes with myoglobin and hemoglobin (Ahn and Maurer, 1990b), and is responsible for producing the pink color (Cornforth et al., 1986; Claus et al., 1994). In this study, the rNIC ratio (ratio of %R537 nm/%R553 nm) was measured as an indicator of nicotinamide hemochrome. The rNIC ratio of the cooked ground chicken products was increased ($p < 0.05$) when 1% NaCl was added, but decreased with higher concentrations of NaCl ($p < 0.05$; Table 4). The rNIC ratio of samples treated with 2% NaCl was not different ($p > 0.05$) from those not treated with NaCl. Samples treated with 3% NaCl showed the lowest rNIC ratio. Overall, the rNIC showed a trend similar to the CIE a^* values in current study. These results indicate that nicotinamide hemochrome may contribute to the development of pink color in fully cooked ground chicken breasts without the addition of pink-generating ligands, if the ground meat is presalted and stored for 7 d before cooking.

Effects of cooking temperature

The endpoint cooking temperature had limited effects on the color and pigment properties of cooked ground chicken breasts (Table 1).

As expected, the cooking yield decreased as the endpoint cooking temperature increased ($p < 0.05$), but there were no differences ($p > 0.05$) between the samples cooked to 70°C and 75°C (Table 5). Murphy and Marks (2000) found that cooking loss in chicken breasts patties increased as the cooking temperature increased from 23°C to 80°C, which is similar to our results.

The pH values ranged from 6.14 to 6.15 across all cooked products and there were no differences ($p > 0.05$) in the pH values, regardless of internal cooking temperatures (Table 5). A similar finding was reported by Claus and Jeong (2018) for

Table 5. Effects of cooking temperature on cooking yield, pH, oxidation reduction potential (ORP) and residual nitrite contents in ground chicken breasts

Cooking temperatures ¹	Dependent variables			
	Cooking yield (%)	pH	ORP (mV)	Nitrite (ppm)
70°C	97.1 ^A	6.14	-106.7 ^B	0.40 ^B
75°C	96.5 ^A	6.15	-106.6 ^B	0.43 ^{AB}
80°C	95.0 ^B	6.15	-106.2 ^{AB}	0.44 ^A
85°C	93.2 ^C	6.15	-105.4 ^A	0.46 ^A
(SE)	(0.55)	(0.09)	(5.92)	(0.05)

¹ Samples were cooked to an internal temperature of 70, 75, 80, and 85°C, respectively, in a 90°C water bath.

^{A-C} Means within a column with different superscript letters are significantly different ($p < 0.05$).

ORP, oxidation reduction potential; Nitrite, residual nitrite contents; SE, standard error.

ground turkey breast products cooked to 71.1°C, 73.9°C, 76.7°C, and 79.4°C. Generally, the pH value affects the PMD, and increasing the pH may cause incomplete denaturation of myoglobin, forming a pink hemochrome in turkey breast and bratwurst (Trout, 1989; Ghorpade et al., 1992), which can lead to increased redness (Davis and Franks, 1995). Thus, consistent pH values at different cooking temperatures may not have influenced myoglobin denaturation and redness in this study.

As endpoint cooking temperature increased, the ORP also increased ($p < 0.05$; Table 5). Cornforth et al. (1986; 1991) found that pink color developed under reducing conditions. In cooked meat, globin denaturation could cause the exposure of heme, and heme iron is oxidized rapidly by air exposure; thus, relatively strong reducing conditions are necessary to form stable pink complexes (Cornforth et al., 1991).

Residual nitrite contents were generally increased with cooking temperature. When samples were cooked at 80°C and 85°C, the residual nitrite contents were higher than when samples were cooked at 70°C. However, no differences ($p > 0.05$) in residual nitrite contents were found between samples cooked at 75°C and 85°C. Ahn and Maurer (1987) reported that at least 1 ppm of nitrite is required to develop pink color in turkey meat. In this study, as residual nitrite content ranged from 0.40 to 0.46 ppm, it is considered that the residual nitrite content did not affect the color and pigment characteristics of cooked chicken breasts.

No differences ($p > 0.05$) were observed in myoglobin contents and PMD for any of the endpoint cooking temperatures tested (Table 6). Similar results were reported by Ghorpade and Cornforth (1993); when pork meat was heated to final internal temperatures of 65°C and 82°C, myoglobin content and PMD was found to be not affected by the cooking temperature.

Further, the endpoint cooking temperature did not affect nitrosyl hemochrome and total pigments ($p > 0.05$; Table 6). Claus and Jeong (2018) obtained similar results in ground turkey breasts cooked at different internal temperatures.

The CIE L* values tended to increase at high cooking temperatures ($p < 0.05$), but there were no differences between the samples cooked to 80°C and 85°C ($p > 0.05$). Similarly, the CIE b* values increased ($p < 0.05$) with cooking temperature (Table 7). In contrast, the CIE a* values were similar ($p > 0.05$) in all cooked samples, regardless of the cooking temperature (Table 7), although they decreased with cooking temperature. Davis and Franks (1995) reported that high endpoint cooking temperatures reduced redness, but increased lightness and yellowness in broiler thigh meat. Helmke and Froning (1971)

Table 6. Effects of cooking temperature on myoglobin contents, percentage myoglobin denaturation (PMD), nitrosyl hemochrome and total pigments in ground chicken breasts

Cooking temperatures ¹	Dependent variables			
	Myoglobin (cooked, mg/g)	PMD (%)	Nitrosyl hemochrome (ppm)	Total pigments (ppm)
70°C	0.23	80.99	1.52	14.76
75°C	0.22	81.72	1.66	14.69
80°C	0.22	82.02	1.60	14.78
85°C	0.22	81.85	1.60	14.75
(SE)	(0.01)	(0.77)	(0.29)	(0.29)

¹ Samples were cooked to an internal temperature of 70, 75, 80, and 85°C, respectively, in a 90°C water bath. Myoglobin, amount of undenatured myoglobin; PMD, percentage myoglobin denaturation; SE, standard error.

Table 7. Effects of cooking temperature on CIE L*, CIE a*, CIE b*, and rNIC in cooked ground chicken breasts

Cooking temperatures ¹	Dependent variables			
	CIE L*	CIE a*	CIE b*	rNIC
70°C	77.17 ^C	3.76	8.10 ^C	1.007
75°C	77.61 ^B	3.63	8.46 ^B	1.007
80°C	77.98 ^A	3.46	8.68 ^{AB}	1.007
85°C	78.22 ^A	3.27	8.88 ^A	1.006
(SE)	(0.41)	(0.33)	(0.35)	(0.0013)

¹ Samples were cooked to an internal temperature of 70, 75, 80, and 85°C, respectively, in a 90°C water bath.

^{A-C} Means within a column with different superscript letters are significantly different ($p < 0.05$).

rNIC, reflectance estimator of nicotinamide hemochrome (%R537 nm/%R553 nm); SE, standard error.

found that lightness was increased, but redness and yellowness were decreased, as temperature increased from 60°C to 82°C in turkey meat, which was partially different from our results.

The rNIC ratio of cooked ground chicken breast samples showed significant difference ($p > 0.05$) among the different treatments. Cornforth et al. (1986) reported that reduced globin or nicotinamide hemochromes could cause the pink color in cooked turkey rolls and that the pigments could be promoted by reducing conditions. Further, high cooking temperatures could lead to a more reducing environment in meat, which is more favorable for the formation of pink hemochromes such as nicotinamide hemochrome (Claus et al., 1994; Ghorpade and Cornforth, 1993; Hamm and Hofmann, 1965; Trout, 1989). However, in our study, the inherent levels of nicotinamide present in ground chicken meat did not affect the rNIC ratio, regardless of the cooking temperature.

Interaction effects of NaCl concentration and endpoint cooking temperature

Interaction effects between NaCl concentration and endpoint cooking temperature were found on cooking yield ($p < 0.0001$) and total pigments ($p < 0.05$) in cooked ground chicken breasts (Table 1). As expected, the cooking yield was lowest ($p < 0.05$) in the samples without NaCl and cooked to 85°C (Table 8). The cooking yield decreased ($p < 0.05$) as the cooking temperature increased. In general, the cooking yield from poultry products has been shown to be decreased when cooking temperature is increased (Claus and Jeong, 2018; Sammel and Claus, 2003; Slesinski et al., 2000b). However, higher levels of NaCl in ground chicken breast had little effect on the cooking yield at the different endpoint temperatures (Table 8). Samples cooked to 85°C had lower ($p < 0.05$) cooking yield than those cooked to lower temperatures.

In samples without NaCl, the total pigments increased ($p < 0.05$) with increasing cooking temperatures. However, samples treated with NaCl did not show significant differences ($p > 0.05$) in total pigments when cooked to different temperatures. Furthermore, when the samples were cooked to 70°C and 75°C, there were no significant differences ($p > 0.05$) in total pigments depending on NaCl concentration. However, the total pigments was higher in samples without salt (0% NaCl) than in those with salt (1%, 2%, and 3% NaCl) when cooked to 80°C and 85°C ($p < 0.05$).

Conclusion

In conclusion, the addition of 0%, 1%, and 2% NaCl had similar effect on the development of pink color in cooked ground chicken breasts, when the meat was presalted for 7 d. The addition of 1% NaCl to ground chicken meat increased the

Table 8. Interaction effects of NaCl concentration and cooking temperature on cooking yield, total pigments in cooked ground chicken breasts

Cooking temperatures ^{1,2}	Dependent variables									
	Cooking yield (%)					Total pigments (ppm)				
	0% NaCl	1% NaCl	2% NaCl	3% NaCl	(SE)	0% NaCl	1% NaCl	2% NaCl	3% NaCl	(SE)
70°C	93.2 ^{Ay}	98.1 ^{Ax}	98.5 ^{Ax}	98.5 ^{Ax}	(0.55)	14.96 ^B	14.85	15.02	14.22	(0.29)
75°C	91.6 ^{By}	97.6 ^{Ax}	97.9 ^{ABx}	98.8 ^{Ax}	(0.55)	14.90 ^B	14.62	14.62	14.62	(0.29)
80°C	88.4 ^{Cy}	96.8 ^{ABx}	96.7 ^{Bx}	98.1 ^{Ax}	(0.55)	15.98 ^{Ax}	14.34 ^y	14.22 ^y	14.56 ^y	(0.29)
85°C	84.7 ^{Dy}	95.3 ^{Bx}	96.5 ^{Bx}	96.2 ^{Bx}	(0.55)	16.26 ^{Ax}	14.05 ^y	14.56 ^y	14.11 ^y	(0.29)
(SE)	(0.55)	(0.55)	(0.55)	(0.55)		(0.29)	(0.29)	(0.29)	(0.29)	

¹ NaCl concentration: all raw ground meat were salted (0, 1, 2, and 3% NaCl, respectively) and stored for 7 d before being cooked.

² Cooking temperature: samples were cooked to an internal temperature of 70, 75, 80, and 85°C, respectively, in a 90°C water bath.

^{A-D} Means within a column with different superscript letters are significantly different ($p < 0.05$).

^{x-y} Means within a row with different superscript letters are significantly different ($p < 0.05$).

SE, standard error.

pinkness of cooked products, along with more formation of nicotinamide hemochrome. However, the addition of 3% salt in ground chicken breasts had a reducing effect on the pigment characteristics associated with the pink color. However, the endpoint cooking temperature had limited effect on the color and pigment characteristics of cooked ground chicken products. Thus, adding less than 2% salt to meat and storing for 7 d can naturally generate a pink color in cooked chicken products. To utilize these practices in the industrial production of meat products, further studies should focus on the mechanisms behind the development of a pink color in meat products, including processing conditions such as storage time and temperature, the type of salt and phosphate used, and other added ingredients.

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References

- Ahn DU, Maurer AJ. 1987. Concentration of nitrate and nitrite in raw turkey breast meat and the microbial conversion of added nitrate to nitrite in tumbled turkey breast meat. *Poult Sci* 66:1957-1960.
- Ahn DU, Maurer AJ. 1989a. Effects of added nitrite, sodium chloride, and phosphate on color, nitrosoheme pigment, total pigment, and residual nitrite in oven-roasted turkey breast. *Poult Sci* 68:100-106.
- Ahn DU, Maurer AJ. 1989b. Effects of added pigments, salt, and phosphate on color, extractable pigment, total pigment, and oxidation-reduction potential in turkey breast meat. *Poult Sci* 68:1088-1099.
- Ahn DU, Maurer AJ. 1989c. Effects of sodium chloride, phosphate, and dextrose on the heat stability of purified myoglobin,

- hemoglobin, and cytochrome c. *Poult Sci* 68:1218-1225.
- Ahn DU, Maurer AJ. 1990a. Poultry meat color: kinds of heme pigments and concentrations of the ligands. *Poult Sci* 69:157-165.
- Ahn DU, Maurer AJ. 1990b. Poultry meat color: pH and the heme-complex forming reaction. *Poult Sci* 69:2020-2050.
- Antonini E, Brunori M. 1971. Hemoglobin and myoglobin and their reaction with ligands. North-Holland Publishing Co., Amsterdam, the Netherlands.
- AOAC. 2007. Official methods of analysis of AOAC International. 18th ed. AOAC International. Gaithersburg, MD, USA. p 931.
- Bernofsky C, Fox JB, Schweigert BS. 1959. Biochemistry of myoglobin. VII. The effect of cooking on myoglobin in beef muscle. *J Food Sci* 24:339-343.
- Claus JR, Jeong JY. 2018. Processing conditions and endpoint temperature effects on development of pink defect without pink-generating ligands in cooked ground turkey breast. *Poult Sci* 97:667-675.
- Claus JR, Shaw DE, Marcy JA. 1994. Pink color development in turkey meat as affected by nicotinamide, cooking temperature, chilling rate, and storage time. *J Food Sci* 59:1283-1285.
- Cornforth DP, Calkins CR, Faustman C. 1991. Methods for identification and prevention of pink color in cooked meat. *Proc Recip Meat Conf* 44:53-58.
- Cornforth DP, Vahabzadeh F, Carpenter CE, Bartholomew DT. 1986. Role of reduced hemochromes in pink color defect of cooked turkey rolls. *J Food Sci* 51:1132-1135.
- Davis CE, Franks DL. 1995. Effect of end-point temperature and storage time on color and denaturation of myoglobin in broiler thigh meat. *Poult Sci* 74:1699-1702.
- Friesen KE, Marcy JA. 2000. Pinking in poultry symposium. 53rd Annual Reciprocal Meat Conference, American Meat Science Association, Columbus, OH, USA. pp 110-112.
- Froning GW, Daddario J, Hartung TE, Sullivan TW, Hill RM. 1969a. Color of poultry meat as influenced by dietary nitrates and nitrites. *Poult Sci* 48:668-674.
- Froning GW, Mather FB, Daddario J, Hartung TE. 1969b. Effect of automobile exhaust fume inhalation by poultry immediately prior to slaughter on color of meat. *Poultry Sci* 48:485-487.
- Ghorpade VM, Cornforth DP. 1993. Spectra of pigments responsible for pink color in pork roasts cooked to 65 or 82°C. *J Food Sci* 58:51-52, 89.
- Ghorpade VM, Cornforth DP, Sisson DV. 1992. Inhibition of red discoloration in cooked, vacuum packaged bratwurst. *J Food Sci* 57:1053-1055.
- Girard B, Vanderstoep J, Richards J. 1989. Residual pinkness in cooked turkey and pork muscle. *Can Inst Food Sci Technol J* 22:372-377.
- Girard B, Vanderstoep J, Richards JF. 1990. Characterization of the residual pink color in cooked turkey breast and pork loin. *J Food Sci* 55:1249-1254.
- Hamm R, Hofmann K. 1965. Changes in the sulphhydryl and disulphide groups in beef muscle proteins during heating. *Nature* 207:1269-1271.
- Heaton KM, Cornforth DP, Moiseev IV, Egbert WR, Carpenter CE. 2000. Minimum sodium nitrite levels for pinking of various cooked meats as related to use of direct or indirect-dried soy isolates in poultry rolls. *Meat Sci* 55:321-329.
- Helmke A, Froning GW. 1971. The effect of end-point cooking temperature and storage on the color of turkey meat. *Poult Sci*

50:1832-1836.

- Holownia K, Chinnan MS, Reynolds AE, Koehler PE. 2003. Evaluation of induced color changes in chicken breast meat during simulation of pink color defect. *Poult Sci* 82:1049-1059.
- Hornsey HC. 1956. The color of cooked cured pork. I. Estimation of the nitric oxide-haem pigments. *J Sci Food Agric* 7:534-540.
- Howe J, Gullett E, Osborne W. 1982. Development of pink color in cooked pork. *Can Inst Food Sci Technol J* 15:19-23.
- Jeong JY. 2017. Effects of short-term presalting and salt level on the development of pink color in cooked chicken breasts. *Korean J Food Sci An* 37:98-104.
- John L, Cornforth D, Carpenter CE, Sorheim O, Pettee BC, Whittier DR. 2005. Color and thiobarbituric acid values of cooked top sirloin steaks packaged in modified atmospheres of 80% oxygen, or 0.4% carbon monoxide, or vacuum. *Meat Sci* 69:441-449.
- Krzywicki K. 1979. Assessment of relative content of myoglobin, oxymyoglobin and metmyoglobin at the surface of beef. *Meat Sci* 3:1-10.
- Ledward DA. 1971. On the nature of cooked meat hemoprotein. *J Food Sci* 36:883-888.
- Lopez K, Schilling MW, Armstrong TW, Smith BS, Corzo A. 2012. Sodium chloride concentration affects yield, quality, and sensory acceptability of vacuum-tumbled marinated broiler breast fillets. *Poult Sci* 91:1186-1194.
- Min B, Cordray JC, Ahn DU. 2009. Effect of NaCl, myoglobin, Fe (II), and Fe (III) on lipid oxidation of raw and cooked chicken breast and beef loin. *J Agric Food Chem* 58:600-605.
- Murphy RY, Marks BP. 2000. Effect of meat temperature on proteins, texture, and cook loss for ground chicken breast patties. *Poult Sci* 79:99-104.
- Nam KC, Ahn DU. 2002. Carbon monoxide-heme pigment is responsible for the pink color in irradiated raw turkey breast meat. *Meat Sci* 60:25-33.
- Puolanne EJ, Ruusunen MH, Vainionpää JI. 2001. Combined effects of NaCl and raw meat pH on water-holding in cooked sausage with and without added phosphate. *Meat Sci* 58:1-7.
- Rhee KS, Ziprin YA. 2001. Pro-oxidant effects of NaCl in microbial growth-controlled and uncontrolled beef and chicken. *Meat Sci* 57:105-112.
- Sackett BAM, Fronting GW, Deshazer JA, Struwe FJ. 1986. Effect of gaseous preslaughter environment on chicken broiler meat quality. *Poult Sci* 65:511-519.
- Sammel LM, Claus JR. 2003. Whey protein concentrates effects on pink color development in a cooked ground turkey breast model system. *Meat Sci* 65:1293-1299.
- Sammel LM, Claus JR. 2006. Citric acid and sodium citrate effects on pink color development of cooked ground turkey irradiated pre- and post-cooking. *Meat Sci* 72:567-573.
- Sammel LM, Claus JR. 2007. Calcium chloride and tricalcium phosphate effects on the pink color defect in cooked ground and intact turkey breast. *Meat Sci* 77:492-498.
- SAS. 2012. SAS/STAT® Software for PC. Release 9.4 versions. SAS Institute Inc., Cary, NC, USA.
- Schwarz SJ, Claus JR, Wang H, Marriott NG, Graham PP. 1998. Quantification of nicotinamide hemochrome in cooked, uncured turkey by reflectance spectrophotometry. *J Muscle Foods* 9:101-110.
- Schwarz SJ, Claus JR, Wang H, Marriott NG, Graham PP, Fernandes CF. 1997. Inhibition of pink color development in cooked, uncured ground turkey through the binding of non-pink generating ligands to muscle pigments. *Poult Sci*

76:1450-1456.

- Schwarz SJ, Claus JR, Wang H, Marriott NG, Graham PP, Fernandes CF. 1999. Inhibition of pink color development in cooked, uncured turkey breast through ingredient incorporation. *Poult Sci* 78:255-266.
- Sebranek JG. 2009. Basic curing ingredients. In *Ingredients in meat products*. Tarté R (ed). Springer Science + Business Media, LLC, New York, NY, USA. pp 1-23.
- Sebranek JG, Fox JB. 1991. Rate of nitric oxide formation from nitrite as affected by chloride ion concentration. *J Muscle Foods* 2:11-20.
- Slesinski AJ, Claus JR, Anderson-Cook CM, Eigel WE, Graham PP, Lenz GE, Noble RB. 2000a. Ability of various dairy proteins to reduce pink color development in cooked ground turkey breast. *J Food Sci* 65:417-420.
- Slesinski AJ, Claus JR, Anderson-Cook CM, Eigel WE, Graham PP, Lenz GE, Noble RB. 2000b. Response surface methodology for reduction of pinking in cooked turkey breast mince by various dairy protein combinations. *J Food Sci* 65:421-427.
- Stewart MR, Zipser MW, Watts BM. 1965. The use of reflectance spectrophotometry for the assay of raw meat pigments. *J Food Sci* 30:464-469.
- Suman SP, Joseph P. 2014. Chemical and physical characteristics of meat: Color and pigment. In *Encyclopedia of meat sciences*. 2nd ed. vol. 3. Dikeman M, Devine C. (ed). Elsevier, Oxford, United Kingdom. pp 244-251.
- Trout GR. 1989. Variation in myoglobin denaturation and color of cooked beef, pork, and turkey meat as influenced by pH, sodium chloride, sodium tripolyphosphate, and cooking temperature. *J Food Sci* 54:536-548.
- Warriss PD. 1979. The extraction of heme pigments from fresh meat. *J Food Technol* 14:75-80.