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Induced Red Discoloration of Broiler Breast Meat: i. Effect of Blood, Bone Marrow and Marination

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Abstract: The bloody, undercooked appearance of fully cooked chicken causes complaints and product rejection by consumers. This defect has been described as a persistent problem with bone-in chicken. Many studies have addressed pink meat or bone darkening, but none have studied the red discoloration problem. Therefore, constituents found in the broiler carcass (breast meat, blood and bone marrow) were combined in an external system to determine the effect of blood and marrow on inducing red discoloration. Three replicate trials were conducted where broiler breast meat was combined with: nothing (control); blood; bone marrow; or, both. An identical set of samples was prepared with added marinade (water, salt and phosphate). Duplicates of each treatment were prepared, placed in glass tubes and cooked. CIE L* (lightness) and a* (redness) values were determined on raw preparations and on cooked meat. Blood, marrow and the combination of both produced significantly ($P < 0.05$) darker and redder raw and cooked breast meat. Blood contributed more to the darkness of raw meat, while marrow contributed more to the darkness of cooked meat and to the redness of both raw and cooked meat. The blood-marrow combination produced darker raw and cooked meat than either ingredient alone, but the combination did not produce redder meat than marrow alone. Marination resulted in darkened raw breast meat, but had little effect on meat darkness or redness when blood, marrow, or both were added. Marrow was determined to be the most important component for inducing red discoloration of breast meat.

Key words: Red discoloration, meat redness, blood, bone marrow, marination

Introduction

Many factors have been shown to affect poultry meat color. Most of the previous research has reported on the deleterious appearance of a diffused pink meat color, which consumers perceive as undercooked. There have been many reports on causes of pinking in poultry meat, summarized by several reviews (Mugler and Cunningham, 1972; Maga, 1994; Froning, 1995). The variety of research into causes of pinking ranges from bird age; diet (intake of nitrites or moldy feed materials); preslaughter transport and handling (stress, automotive exhaust, mishandling and improper stunning); and processing and cooking methods including meat aging, nitrate or nitrite contamination and cooking equipment (Froning and Hartung, 1967; Froning *et al*., 1969a; Wu *et al*., 1994; Froning *et al*., 1978; Froning *et al*., 1969b; Walker *et al*., 1993; Bilgili, 1992; Young *et al*., 1996; Nash *et al*., 1985; Heath and Owens, 1992; Cornforth *et al*., 1998). Some of these research projects exposed birds or meat to conditions or compounds that had been identified as causative agents of meat pinking. Other researchers have expanded on this idea and have induced pinking in laboratory situations to both understand the mechanisms causing pinking and to test methods or ingredients that prevent or reduce pinking (Schwarz *et al*., 1999; Slesinski *et al*., 2000; Holownia *et al*., 2003).

Bone darkening is a type of color defect found in cooked chicken meat, where the tissue around the bone is discolored with a burgundy or black appearance. This defect was first associated with frozen poultry (Koonz and Ramsbottom, 1947; Brant and Stewart, 1950; Spencer *et al*., 1961). The dark discoloration is apparently a result of bone marrow leaching through the bone onto surrounding meat, which then becomes dark when cooked. Cooking method has been reported to affect bone darkening, where freezing prior to cooking increased the severity of darkening more than cooking followed by freezing and reheating (Lyon and Lyon, 1986). Blast freezing increased darkness and redness of raw and cooked bone-in broiler thighs, while removal of the femur prior to freezing decreased the redness of raw thighs (Lyon *et al*., 1976).

Another type of color problem that affects poultry meat is the intense red, bloody, localized discoloration of bone-in fully-cooked product. A previous report showed that 11% of several different cooked chicken products available at retail were either severely or extensively affected by red discoloration (Smith and Northcutt, 2003). Less research has been conducted on this type of discoloration than other meat color problems, probably due to the sporadic nature of occurrences during production. No reports on the inducement of this type of discoloration for laboratory study were found.

Marination has been shown to affect meat color. Increasing the levels of phosphate marination resulted in increased darkness of cooked ground chicken (Froning, 1966). Trisodium phosphate marinade was also reported to reduce the lightness of the exterior and interior of cooked chicken breast fillets (Yang and Chen, 1993). Redness of cooked chicken breast meat (Young *et al.*, 1996; Young and Lyon, 1997) and raw breast meat (Lyon *et al.*, 1998) was reduced by phosphate marination. Conflicting reports have also been published that show marination either had no effect on chicken breast meat color (Young *et al.*, 1999), increased lightness of cooked chicken breast meat (Young *et al.*, 1996), or increased redness of early deboned turkey breast (Young and Lyon, 1994).

The development of a model system that would consistently produce red discolored broiler meat, composed only of naturally occurring poultry tissue components, would be beneficial as a research tool for conducting efforts to minimize or eliminate this problem. Primarily the objective of this experiment was to attempt to create a system that would consistently produce red discoloration by introducing large quantities of natural constituents (blood, bone marrow, or both) to broiler breast meat. Marinade was also included to determine its potential role in meat discoloration.

Materials and Methods

Boneless, skinless broiler breast meat and whole broiler thighs were obtained fresh (refrigerated) from a local retail store. Both breast meat and thighs were refrigerated at 4°C until used later that day. Breast meat was removed from refrigeration, trimmed of fat and connective tissue, manually chopped into pieces and blended for 30 s in a food processor.

Thighs were removed from refrigeration, manually deboned, then femurs were cleaned of excessive external tissue, both epiphyseal end caps were removed and the diaphysis was cut longitudinally to allow removal of the fatty, semi-liquid marrow with a narrow spatula. The marrow was manually mixed, then blended for 10 s in a food processor to a thick liquid immediately prior to use.

Chicken blood was obtained fresh from a 6 week old broiler killed via exsanguination approximately 1 h prior to the start of each trial. The fresh clotted blood was placed in a food processor for 10 s and homogenized to a viscous liquid immediately prior to use.

Breast meat, bone marrow and blood were manually mixed together for one minute in the following combinations (Table 1): 10 g meat only (control); meat plus 1 g blood; meat plus 1 g marrow; meat plus 1 g blood and 1 g marrow. Additionally, 1 g of a marinade mixture (92% water, 5% sodium chloride and 3% sodium polyphosphates) was added to 10 g breast meat, manually mixed, then blood, marrow, or both were added as previously described. Duplicates of each of these seven treatments plus control were prepared. A colorimeter (Minolta Chroma Meter 300, Minolta Corp., Ramsey, NJ 07446) was used to record CIE L* and a* values in triplicate from each of three separate locations of each prepared mixture.

Table 1: Treatments showing the possible combinations of ingredients (1 g each of blood, marrow, or marinade) added to 10 g of raw broiler breast meat, then cooked

Treatment	Blood	Marrow	Marinade
	----------(1 g each)----------		
Control (10 g meat)	-	-	-
Blood	+	-	-
Marrow	-	+	-
Blood+Marrow	+	+	
Marinade only	-	-	+
Marinade+Blood	+	-	+
Marinade+Marrow	-	+	+
Marinade+Blood+Marrow	+	+	+

n=6

Each of the seven mixtures plus the control (in duplicate) was placed into one end of glass tube (OD = 20 mm, ID = 17 mm X 200 mm length) and sealed on the bottom with a # 2 rubber stopper. Prepared tubes were placed in a non-circulating water bath set at 95°C for approximately 45 minutes, until an internal temperature of 74°C was reached as determined by an inserted thermocouple. Tubes were then immediately placed in an ice water bath for 1 h. Stoppers were removed, the cooked mixture "plugs" were taken from the tubes and sliced longitudinally 3 times with a razor blade, producing "coins" with a flat surface that was approximately 5 mm thick and 16 mm in diameter. L* (lightness) and a* (redness) values were determined (triplicate readings averaged together) for each of the three slices per plug using the colorimeter.

Three replicate trials were conducted, with duplicates prepared for each of the seven treatments and control in each trial; three separate areas of each of the prepared raw mixtures and three separate slices per tube of each mixture were measured, resulting in a 3 X 8 X 2 X 3 design and a total sample number of 144 (each raw and cooked). The three slices per plug were not averaged together per tube as the mixtures within the tube were not completely homogenous. Data were pooled together across trials and duplicates within trial and analyzed for differences between treatments using SAS software general linear models procedures (SAS Institute, 1999). Main effects tested were trial, treatment and any effects of duplicate or area within sample. Means were separated using Duncan's multiple range test ($P < 0.05$). Data were pooled across trials and residual error was used as the test statistic even where trial by treatment interactions were significant, because the

L*and a* of the original ingredients (meat, blood and marrow) varied by trial. For example, raw breast meat varied in L* from 57.4 to 59.7 to 63.6 and for a* from 0.5 to 3.8 to 4.7 in trials one, two and three, respectively. Similar variations were observed for the raw blood and marrow. However, these differences stayed in the same pattern within each trial and across trials the pattern was the same, where breast meat was darkened and reddened by addition of blood, marrow, or both, whether raw or cooked. Therefore the trial by treatment interaction was considered an artifact from natural variation of the beginning ingredients and not due to differences in experimental technique between trials.

Results

The effect of blood, bone marrow, both and added marinade on L* (lightness) and a* (redness) of raw broiler breast meat is shown in Table 2. The control lightness value was 60.20, which was significantly ($P < 0.05$) darkened to 46.15 by marrow, then further reduced to 44.55 by blood and then dropped to the lowest value of 41.88 by the blood-marrow combination. Adding marinade darkened meat (57.82) as compared to the control value (60.20). Blood and marrow significantly increased darkness values of marinaded meat (45.15 and 45.02, respectively), while the combination lowered values further (41.55). Redness values were significantly increased from the control value (3.67) by blood (15.01), while marrow and the blood marrow combination produced even higher redness values (17.88 and 17.36, respectively). Marinade addition had no effect on redness values as compared to the control (3.06 vs. 3.67). Blood significantly increased marinaded meat redness values to 14.45, while marrow and the blood-marrow combination produced the highest redness values of 17.31 and 16.83, respectively, which were not different from each other. The blood-marrow combination darkened raw meat more than either component alone, but that effect may have been due to the extra 1 g of constituent rather than a combined chemical effect. The blood-marrow combination also produced redder meat than blood alone, but meat was not redder than marrow alone. Overall, blood seemed to have the most effect on decreasing lightness values and marrow the most effect on increasing redness values of raw breast meat. Marinade addition slightly lowered the lightness value of breast meat, but had no effect on either lightness or redness values of breast meat when blood, marrow, or both were added.

The effect of blood, marrow and marinade on L* and a* values of cooked broiler breast meat are shown in Table 3. The control lightness value (81.39) was significantly darkened by blood (60.71), lowered further by marrow (58.62) and then reached the lowest value from the blood-marrow combination (54.10). Adding marinade had no effect on lightness values as compared to the control (79.70 vs. 81.39, respectively). Blood lowered lightness values to 61.06, while marrow further lowered values to 57.10 and the blood-marrow combination produced the lowest value at 53.44. The redness value for the control was 0.02, which was increased significantly to 5.76 by blood, then further increased to 7.10 and 7.48 by marrow and the blood-marrow combination, respectively. The redness value for marinaded meat (-0.37) was not different from control (0.02). Blood significantly increased redness values to 4.66, marrow increased redness to 6.31 and the blood-marrow combination further increased redness to 7.21. As observed with raw meat, the blood-marrow combination produced darker and redder cooked meat than either blood or marrow alone. The only exception was non-marinaded meat redness, where the blood-marrow combination value (7.48) was not different than marrow alone (7.10). Generally marinade had no effect on lightness or redness cooked meat values, although redness from added blood was lowered slightly. Non-marinaded meat blood values of 5.76 dropped to 4.66 for marinaded meat. Marrow produced darker and redder

Table 2: Lightness (L*) and redness (a*) values[1] (means ± SEM) of raw broiler breast meat (control) with added blood, marrow, a combination of both and the same mixtures with added marinade

	L*	a*
Control	60.20^{a}±0.63	3.67^{c}±0.25
Blood	44.55^{d}±0.44	15.01^{b}±0.72
Marrow	46.15^{c}±0.70	17.88^{a}±1.02
Blood+Marrow	41.88^{e}±0.43	17.36^{a}±0.75
Marinade only	57.82^{b}±0.60	3.06^{c}±0.32
Marinade+Blood	45.02^{d}±0.54	14.45^{b}±0.69
Marinade+Marrow	45.15^{cd}±0.56	17.31^{a}±0.75
Marinade+Blood+Marrow	41.55^{e}±0.57	16.83^{a}±0.89

[1]n=18. [a-e]Means in columns with no common superscripts are significantly different ($P<0.05$)

Table 3: Lightness (L*) and redness (a*) values[1] (means ± SEM) of cooked broiler breast meat (control) with added blood, marrow, a combination of both and the same mixtures with added marinade

	L*	a*
Control	81.39^{a}±0.21	0.02^{e}±0.14
Blood	60.71^{b}±0.69	5.76^{c}± 0.32
Marrow	58.62^{c}±1.13	7.10^{ab}± 0.54
Blood+Marrow	54.10^{d}±0.61	7.48^{a}±0.32
Marinade	79.70^{a}±0.26	-0.37^{e}± 0.18
Marinade+Blood	61.06^{b}±0.89	4.66^{d}±0.43
Marinade+Marrow	57.10^{c}±0.77	6.31^{bc}± 0.45
Marinade+Blood+Marrow	53.44^{d} ±0.91	7.21^{a}± 0.44

[1]n=18. [a-e]Means in columns with no common superscript are significantly different ($P<0.05$)

cooked broiler breast meat than blood. The total difference between cooked meat control values and either blood or marrow showed that marrow contributed more to lower lightness and higher redness values than did blood. This same pattern was evident for raw meat redness values. Therefore marrow was considered overall to be more important to contributing to red discoloration than blood.

Discussion

Excess blood in or on broiler meat is possible during normal processing conditions. Electrical stunning, as practiced for humane slaughter on the vast majority of broilers processed in the US, has been found to result in a higher incidence of hemorrhagic leg syndrome scores indicated by excessive blood around the femur (Walker *et al.*, 1993). An alternative, whole body stun method for broiler chickens was reported to produce even more muscle hemorrhages than head-only stunning (Kranen *et al.*, 1996). Meat that had been bruised, a common defect due to mishandling prior to slaughter, would also contain enough excess blood to discolor the muscle. Although the amount of blood introduced to breast meat in this study was excessive, the CIE L* value from added blood was similar to the L* value reported by Northcutt *et al.* (2000) for newly bruised raw broiler breast meat (44.93 vs. 44.55, respectively). The a* value in that same prior study for newly bruised tissue was lower than that reported in this study for raw broiler breast meat with added blood, however (8.38 vs. 15.01, respectively). Overall, blood was shown to be more important than marrow for contributing to the dark discoloration of broiler breast meat.

Bone marrow is obviously present in bone-in chicken, but typically is not in contact with muscle tissue. Miscuts through the bone during processing are possible and could lead to the introduction of marrow onto meat surfaces (Smith and Northcutt, 2003). Leaching of the marrow after freezing has been shown with previous bone darkening research, but the leached marrow exhibits a black appearance when cooked (Koonz and Ramsbottom, 1947; Brant and Stewart, 1950; Spencer *et al.*, 1961; Lyon and Lyon, 1986). Freezing may inactivate the factor or factors involved with producing redness of non-frozen meat. Removal of the femur from thighs prior to cooking and subsequent decreased redness in cooked meat indicates the femur and presumably the marrow, does affect meat redness (Lyon *et al.*, 1976). The CIE a* values reported in a previous study for discolored areas of commercial cooked breast meat products ranged from 4.15 to 8.07, which was similar to the cooked meat with added marrow a* value of 6.31 and 7.10 (with and without added marinade, respectively) reported in this study (Smith and Northcutt, 2003). Marrow contributed more to the red discoloration of meat, whether raw or cooked, than did blood in this study.

Marination darkened raw breast meat, but otherwise had little effect on discoloration introduced by blood, bone marrow, or the blood-marrow combination. Previous researchers have reported a variety of sometimes contradicting results concerning breast meat color and marination. Marination has been shown to have no effect on normal breast might lightness or redness whether raw (Yang and Chen, 1993; Young *et al.*, 1996; Northcutt *et al.*, 2000) or cooked (Lyon *et al.*, 1998; Young *et al.*, 1999; Northcutt *et al.*, 2000). Similarly, another report showed no effect of marination on just the lightness of raw meat (Lyon *et al.*, 1998). Others researchers have reported marination increased lightness of cooked meat (Young *et al.*, 1996), but Yang and Chen (1993) found marination decreased lightness of cooked breast meat, which was the same result Froning (1966) showed for cooked ground chicken. Marination increased redness in raw (Lyon *et al.*, 1998) or cooked (Yang and Chen, 1993) breast meat, but others reported marination decreased redness in cooked breast meat (Young *et al.*, 1996; Young and Lyon, 1997). Also, Northcutt *et al.* (2000) found marination increased the redness of raw breast meat but decreased the redness of cooked breast meat that had been recently bruised. The lack of effect produced by marination in the present study was probably due to the relatively excessive amounts of added blood and bone marrow, which prevented marinade from affecting meat color to any measurable extent.

The current study provides evidence that discoloration (both darker and redder than normal) was induced in broiler breast meat. Although the addition of blood, bone marrow, or both in large quantities relative to the amount of muscle tissue did produce darker and redder cooked meat, the discoloration was a burgundy (reddish-brown) or darker color. This color was very similar to the descriptions previously provided for bone darkening (Koonz and Ramsbottom, 1947; Brant and Stewart, 1950). Cooked meat exhibiting intense red color and bloody juice was not produced, except for one slice of one plug from the added marrow treatment. This slice did exhibit intense redness in the meat and a small amount of bloody juice. The only difference was that slice contained a small piece (approximately 3 mm) of bony marrow that was inadvertently left in the sample. This finding, plus the ability of bone marrow more than blood to redden meat, will result in a further testing and possible modification of this model system for inducing red discoloration in broiler breast meat.

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