EFFECTS OF PACKAGING ON BONE MARROW DISCOLORATION IN BEEF ARM, RIB, SHOULDER BLADE, AND THORACIC VERTEBRA BONES

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Summary

Meat retailers have reported bone marrow discoloration to be a problem, especially in modified-atmosphere packages (MAP). To evaluate causes of bone marrow discoloration in different beef bones and packaging systems, 36 beef arm bones, ribs, shoulder blades, and thoracic vertebrae from USDA Select and Choice carcasses were obtained from a commercial abattoir, cut into 1-inch-thick sections at 4 days postmortem, and packaged into 1) polyvinyl chloride film (PVC) overwrap; 2) high-oxygen (80% O_2, 20% CO_2) MAP; or 3) ultra-low-oxygen (70% N_2, 30% CO_2) MAP. Packages were displayed under continuous fluorescent lighting for 4 days at 35.6˚F. Ribs, shoulder blades, and thoracic vertebrae packaged in PVC and high-oxygen MAP developed undesirable gray or black discoloration during display. In ultra-low-oxygen MAP, mean visual-color scores were acceptable throughout display. The a* values (larger values equate to redder color) for ribs, shoulder blades, and thoracic vertebrae decreased ($P<0.05$) over time. Arm-bone marrow had less oxidation and dramatically less total iron and hemoglobin than did marrow from ribs and thoracic vertebrae. The much larger amounts of iron and hemoglobin in ribs and thoracic vertebrae likely correspond to marrow discoloration. In summary, bone marrow discoloration occurs in ribs, shoulder blades, and thoracic vertebrae packaged in PVC or high-oxygen MAP. Bones packaged in ultra-low-oxygen MAP or arm bones packaged in PVC or high-oxygen MAP had minimal oxidation and discoloration.

Introduction

Occurrence of the 'black bone' condition in modified-atmosphere packages (MAP) of bone-in, beef retail cuts has been reported by meat retailers. Consumers may perceive bone discoloration ('black bone') as unwholesome, and it may affect their overall perception of a fresh meat product. Bone marrow discoloration has been reported in high-oxygen MAP beef and pork and also in cuts packaged in polyvinyl chloride film (PVC). As more meat is being sold as case-ready, it is important to find causes of, and preventions for, this problem.

One researcher suggested that bone blackening occurs when bone is cut and hemoglobin is released to the surface, where it will accumulate when the red blood cells are disrupted. Over time and through exposure to air, hemoglobin on the surface of the bone turns from red to brown to black. Other possibilities include bone marrow that contains more total pigments, more hemoglobin, and more iron when compared with muscle. Furthermore, lipid content in bovine bone marrow differs among bones and among their locations. The lipid contents of bovine bone marrow from cervical vertebrae, lumbar vertebrae, and leg bones have been analyzed. Bone marrow from cervical vertebrae contained the least lipid, whereas marrow from leg bones had the most. In addition, bone marrow resembles adipose tissue more than it resembles muscle or liver tissue. Thus, lipid oxidation may also be a factor in the development of bone marrow discoloration. Beef lumbar vertebrae have been found to discolor within 24 hours when packaged in high-oxygen MAP, primarily due to the oxidation of hemoglobin.

The objectives of this experiment were to determine the prevalence in different packaging systems of bone marrow discoloration in beef arm bones, ribs, thoracic vertebrae, and shoulder blades, and to determine factors that may cause bone marrow discoloration.

Procedures

Thirty-six beef arm bones, ribs, shoulder blades, and thoracic vertebrae from USDA Select and Choice carcasses obtained from a commercial abattoir were cut into 1-inch-thick sections at 4 days postmortem by using a band saw, and sections were packaged into one of three package types: 1) PVC overwrap; 2) high-oxygen (80% O_2, 20% CO_2) MAP; and 3) ultra-low-oxygen (70% N_2, 30% CO_2) MAP. One each of an arm bone, a shoulder blade, and a thoracic vertebra, and two rib bones were placed in each package. The PVC samples were packaged in foam trays with oxygen-permeable film. High-oxygen and ultra-low-oxygen MAP packages were packaged in rigid plastic trays and were covered with barrier lidding film. Each ultra-low-oxygen MAP had one activated oxygen scavenger added to the package. There were two replications of 18 packages of each system. Within each replication, 12 packages remained in the display case through day 4, whereas 6 packages were opened on day 2 of display (mid display) for instrumental color readings.

Packages were displayed under continuous fluorescent lighting for 4 days at 35.6˚F. Packages were rotated twice daily to maintain a random display case placement.

Instrumental CIE a* measurements were collected with a Hunter labscan 2. CIE a* measures red (+) to green (-). Bones were scanned before packaging on day 0 and after visual color scores were collected on days 2 and 4.

Ten trained visual panelists scored the porous portion of bone-marrow for color once each day for 4 days, beginning on day 0. Bone sections in high-oxygen MAP and in PVC packages were scored according to the seven-point scale: 1) bright reddish-pink to red, 2) dull pinkish-red, 3) slightly grayish-pink or grayish-red, 4) grayish-pink or grayish-red, 5) moderately gray, 6) all gray or grayish-black, and 7) black discoloration. Ultra-low-oxygen MAP bones were scored according to the seven-point scale: 1) bright purplish-red or purplish-pink, 2) dull purplish-pink or purplish-red, 3) slightly grayish purple or pink, 4) grayish-purple or grayish-red, 5) moderately gray, 6) all gray or grayish-black, and 7) black discoloration.

Bone marrow was extracted and analyzed for: 1) 2-thiobarbituric acid reactive substances (TBARS) content, a measure of oxidation, 2) myoglobin and hemoglobin pigment concentrations, and 3) total iron and phosphorus concentrations.

Results and Discussion

Visual Color Display. Visual color scores for the four bone types and three packaging systems are shown in Table 1. Arm-bone marrow became darker ($P<0.05$) with each day of display, but by day 4 of display, the visual score was still only a 'dull pinkish-red' for bones packaged in PVC or high-oxygen MAP and a 'slightly grayish purple or pink' for those packaged in ultra-low-oxygen MAP. According to panelist observations, the arm bones did not turn gray or black during display and remained acceptable in color. There were no differences between PVC and high-oxygen packages for arm-bone visual color scores. No comparisons were made between visual color of bones packaged in PVC or high-oxygen MAP with visual color of bones packaged in ultra-low-oxygen MAP because different color scales were used for the packaging methods.

Visual scores for rib-bone marrow increased (more gray) ($P<0.05$) with increased display time in all packaging types. In samples packaged in PVC or high-oxygen MAP, the ribs were 'grayish-pink or -red' by day 1 of display and were 'moderately gray' from day 2 to 4. Ribs packaged in ultra-low-oxygen MAP were only 'slightly grayish purple or pink' by the end of display.

Shoulder-blade bone marrow became darker ($P<0.05$) with increased display time in all three packaging types. Visual color scores for bones packaged in PVC or high-oxygen MAP were 'grayish-pink or -red' beginning on day 1 of display, whereas those in ultra-low-oxygen MAP were only 'slightly grayish-pink or -red'. Visual color scores were lower for bones packaged in PVC than for those packaged in high-oxygen MAP at all days except day 0 and 4.

Visual color scores showed that bone marrow from thoracic vertebrae turned dark ($P<0.05$) by day 1 of display and continued to become darker through day 4 of display. In samples packaged in PVC or high-oxygen MAP, visual scores were already 'moderately gray' by day 1, whereas thoracic vertebra packaged in ultra-low-oxygen MAP were only 'slightly grayish-pink or -red'. Package differences ($P<0.05$) were only found between PVC and high-oxygen MAP on day 0.

In PVC or high-oxygen MAP packaging, arm-bone marrow did not discolor and had visual color scores much more desirable than those of the other bones (approximately a 3-point advantage on a 7-point scale from day 1 through 4 of display). Ribs, shoulder blades, and thoracic vertebrae packaged in PVC or high-oxygen MAP had undesirable discoloration, with a significant proportion described as 'black bone.' In ultra-low-oxygen MAP packaging, mean visual color scores were acceptable throughout display. In addition, arm bones and ribs had better scores (less gray or black) ($P<0.05$) than did shoulder blades and thoracic vertebrae.

Instrumental Color. In general, a* values decreased more (less red) for ribs, shoulder blades, and thoracic vertebrae in PVC or high-oxygen MAP packaging than in ultra-low-oxygen MAP packaging (Table 2). Instrumental a* readings show results similar to those observed by panelists in the visual color scores for bones packaged in PVC or high-oxygen MAP. Changes in color were minor between day 2 and day 4 for bones packaged in ultra-low-oxygen MAP.

Arm bones had smaller a* (less red) values than did the other bones. The a* values for ribs, shoulder blades, and thoracic vertebrae decreased over time, which corresponded to increased visual color scores (more discoloration). In addition, a* value changes from bones packaged in ultra-low-oxygen MAP were smaller, matching much smaller changes in visual color score.

Differences in a* between bones packaged in either PVC or high-oxygen MAP packaging and in ultra-low-oxygen MAP packaging at day 2 and 4 in all bone types would be expected because of the presence or lack of oxygen in the packages, respectively. Initial instrumental color was measured before packaging; therefore, we would expect values at day 0 to be similar, but values at day 2 and 4 to be different, among the different packages.

Bone Marrow Analyses. 2-Thiobarbituric acid reactive substances (TBARS) for bones in different package types during display are shown in Table 3. Oxidation was considerably less for arm-bone marrow than for marrow from ribs and thoracic vertebrae and, for the arm bone, did not change over display time. Ultra-low-oxygen MAP packaging resulted in the least change in TBARS from day 0 to 4.

Marrow from ribs and thoracic vertebrae had dramatically more ($P<0.05$) total iron and hemoglobin than did arm-bone marrow (Table 4). Arm-bone and rib marrow had more ($P<0.05$) phosphorus than did marrow from thoracic vertebrae; rib marrow had more ($P<0.05$) myoglobin than did marrow from thoracic vertebrae (Table 4). Myoglobin was undetectable in arm-bone marrow. The much greater total iron and hemoglobin in ribs and thoracic vertebrae likely corresponds to bone marrow discoloration.

Overall, the ribs, shoulder blades, and thoracic vertebrae turned dark ('grayish-black') in PVC or high-oxygen MAP packaging during 4 days of display. These bones turned dark within 24 hours. Preliminary research within our laboratory showed that this happened between approximately 5 and 24 hours after packaging (data not shown). In contrast, arm bones remained acceptable in color throughout 4 days of display. One possible explanation for arm bones maintaining an acceptable color while ribs, shoulder blades, and thoracic vertebrae darkened over storage time is the difference in bone marrow composition. There are two types of marrow; red and yellow. Red marrow is described as the hemopoietically active marrow that is present in vertebrae and ribs. Yellow marrow is adipose tissue in bone marrow and is found in the distal portion of long bones. Thus, arm-bone marrow contains much more yellow marrow and lacks the abundance of red marrow and hemoglobin found in ribs and vertebrae that show more extreme discoloration. If the major component of arm-bone marrow is lipid, and lipid oxidation shows an increase in TBARS values, then the extremely small TBARS values found in arm-bone marrow in our study indicate that lipid oxidation is not a primary cause of marrow discoloration.

Some change in color occurs in bones packaged in ultra-low-oxygen MAP; overall, however, these bones remain acceptable in color and will bloom to a reddish color when exposed to oxygen. The lack of oxygen in the ultra-low-oxygen MAP may inhibit or greatly slow oxidation in ribs, shoulder blades, and vertebrae. When packaged in PVC or high-oxygen MAP, ribs, shoulder blades, and thoracic vertebrae discolor. The TBARS results suggest that less oxidation of hemoglobin and(or) myoglobin in marrow from thoracic vertebrae occurs in ultra-low-oxygen MAP packaging, compared with that in PVC or high-oxygen MAP packaging.

Bone marrow discoloration occurred in ribs, shoulder blades, and thoracic vertebrae packaged in PVC or high-oxygen MAP. Bones packaged in ultra-low-oxygen MAP, or arm bones packaged in PVC or high-oxygen MAP, had minimal discoloration. It seems likely that bone discoloration is caused primarily by oxidation of hemoglobin, but heme-catalyzed lipid oxidation, or a combination of the two, could play roles.

Table 1. Visual Color Scores[a,b] for Bones in Different Package Types from Day 0 to 4 of Display at 35.6°F

Bone	Package[c]	Day				
		0	1	2	3	4
Arm	PVC	1.5^{v}	2.0^{w}	2.2^{x}	2.3^{y}	2.6^{z}
Arm	High	1.4^{w}	1.8^{x}	2.1^{y}	2.2^{y}	2.5^{z}
Arm	Ultra-low	1.8^{w}	2.5^{x}	2.9^{y}	2.8^{y}	3.0^{z}
Ribs	PVC	1.7^{ev}	4.6^{dw}	5.0^{dx}	5.2^{dy}	5.3^{dz}
Ribs	High	1.4^{dw}	4.6^{dx}	5.1^{dy}	5.2^{dy}	5.3^{dz}
Ribs	Ultra-low	2.1^{w}	2.5^{x}	2.8^{y}	3.0^{z}	3.1^{z}
Shoulder blade	PVC	2.1^{ev}	4.2^{dw}	4.9^{dx}	5.1^{dy}	5.5^{dz}
Shoulder blade	High	1.8^{dv}	4.6^{ew}	5.2^{ex}	5.4^{ey}	5.6^{dz}
Shoulder blade	Ultra-low	2.3^{v}	3.1^{w}	3.4^{x}	3.6^{y}	3.7^{z}
Thoracic vertebra	PVC	2.2^{ew}	5.3^{dx}	5.8^{dy}	5.8^{dy}	6.1^{dz}
Thoracic vertebra	High	1.6^{dw}	5.2^{dx}	5.6^{dy}	5.8^{dyz}	5.9^{dz}
Thoracic vertebra	Ultra-low	2.7^{w}	3.1^{x}	3.3^{y}	3.4^{yz}	3.5^{z}

[a]Standard error for all means = 0.14.

[b]High-oxygen and PVC color scale: 1=bright reddish-pink to red, 2=dull pinkish-red, 3=slightly grayish-pink or grayish red, 4=grayish-pink or grayish red, 5=moderately gray, 6=all gray or grayish-black, and 7=black discoloration; Ultra-low-oxygen color scale: 1=bright purplish-red or purplish-pink, 2=dull purplish-pink or purplish-red, 3=slightly grayish purple or pink, 4=grayish-purple or grayish-red, 5=moderately gray, 6=all gray or grayish-black, 7=black discoloration.

[c]PVC = polyvinyl chloride overwrap film; High = high-oxygen modified-atmosphere packaging; and Ultra-low = ultra-low-oxygen modified-atmosphere packaging.

[d,e]Means with different superscript letters within bone type and within columns (PVC vs. High) differ ($P<0.05$).

[v,w,x,y,z]Means with different superscript letters across rows (days) differ ($P<0.05$).

Table 2. Instrumental CIE a* Values for Bones in Different Package Types from Day 0, 2, and 4 of Display at 35.6°F

Bone	Package[a]	Day		
		0	2	4
Arm	PVC	14.0[z]	15.2[cz]	15.2[cz]
Arm	High	13.7[y]	15.0[cyz]	15.1[cz]
Arm	Ultra-low	13.6[z]	11.3[by]	10.2[by]
Ribs	PVC	26.6[z]	18.1[by]	16.7[by]
Ribs	High	25.7[z]	19.4[by]	17.3[bx]
Ribs	Ultra-low	25.7[z]	22.8[cy]	21.2[cy]
Shoulder blade	PVC	25.1[z]	18.6[by]	16.3[bx]
Shoulder blade	High	24.0[z]	20.2[by]	15.6[bx]
Shoulder blade	Ultra-low	24.6[z]	23.9[cz]	24.0[cz]
Thoracic vertebra	PVC	26.6[z]	16.5[by]	14.4[bx]
Thoracic vertebra	High	25.7[z]	18.7[by]	14.3[bx]
Thoracic vertebra	Ultra-low	26.2[z]	22.6[cy]	22.7[cy]
SEM	--	0.52	0.85	0.62

[a]PVC = polyvinyl chloride overwrap film; High = high-oxygen modified-atmosphere packaging; and Ultra-low = ultra-low-oxygen modified-atmosphere packaging.
[b,c]Means with different superscript letters within bone type and within columns differ P<0.05).
[x,y,z]Means with different superscript letters across rows (days) differ (P<0.05).

Table 3. 2-Thiobarbituric Reactive Substances[a] for Bones in Different Package Types from Day 0 and 4 of Display at 35.6°F

Bone	Package[b]	Day 0	Day 4	SEM
Arm	PVC	0.03[z]	0.06[z]	0.02
Arm	High	0.03[z]	0.06[z]	
Arm	Ultra-low	0.03[z]	0.04[z]	
Ribs	PVC	0.74[z]	0.77[dz]	0.03
Ribs	High	0.74[y]	0.84[dz]	
Ribs	Ultra-low	0.74[y]	0.65[cz]	
Thoracic vertebra	PVC	0.67[y]	1.04[dz]	0.03
Thoracic vertebra	High	0.67[y]	1.01[dz]	
Thoracic vertebra	Ultra-low	0.67[y]	0.75[cz]	

[a]mg malonaldehyde/ kg sample.
[b]PVC = polyvinyl chloride overwrap film; High = high-oxygen modified-atmosphere packaging; and Ultra-low = ultra-low-oxygen modified-atmosphere packaging.
[c,d]Means with different superscript letters within bone types and within columns differ ($P<0.05$).
[y,z]Means with different superscript letters across rows (days) differ ($P<0.05$).

Table 4. Total Iron, Phosphorus, Hemoglobin, and Myoglobin for Bone Marrow from Arm Bones, Ribs, and Thoracic Vertebrae

Bone	Total Iron (ppm)	Phosphorus (ppm)	Hemoglobin (mg/g)	Myoglobin (mg/g)
Arm	8.1[x] (±0.54 [a])	868[z] (±93)	4.5[y] (±0.45)	ND[b]
Ribs	237[z] (±11)	847[z] (±44)	160[z] (±13)	0.530[z] (±0.05)
Thoracic Vertebra	219[y] (±8.9)	574[y] (±42)	153[z] (±6.4)	0.313[y] (±0.02)

[a]Standard error of the mean.
[b]Not detectable.
[x,y,z]Means with different superscript letters within columns differ ($P<0.05$).