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Spoilage characteristics of ground beef with added lactic acid bacteria and rosemary oleoresin packaged in a modified-atmosphere package and displayed at abusive temperatures

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ABSTRACT: Lactic acid bacteria (LAB) can reduce *Escherichia coli* O157:H7 and *Salmonella* spp. in ground beef during storage. Furthermore, the addition of rosemary oleoresin (RO), a natural antioxidant, to ground beef has been shown to increase shelf life and is commonly used in modified-atmosphere packaged (MAP) ground beef. This study evaluated the effects of LAB and RO treatment on the shelf life and stability of MAP ground beef displayed at abusive (10°C) temperatures for 36 h. Subjective and objective sensory analyses were conducted to determine spoilage endpoints. Trained and consumer panel responses and Hunter lightness (L^*), redness (a^*), and yellowness (b^*) values were not affect-

ed ($P = 0.62, 0.66, 0.45$) by LAB addition, although RO inclusion improved ($P < 0.05$) lean color. Ground beef with LAB and RO had significantly less ($P < 0.0001$) thiobarbituric acid reactive substance values than control ground beef, indicating decreased lipid oxidation. Additionally, RO inclusion reduced ($P < 0.0001$) off odors, as determined by trained and consumer odor panels. Overall, the addition of LAB did not negatively affect beef color, odor, or oxidative rancidity, suggesting that LAB can be added to ground beef in MAP packaging as a processing intervention without detrimentally affecting shelf life or stability.

Key words: ground beef, lactic acid bacteria, modified-atmosphere packaging, spoilage, temperature abuse

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INTRODUCTION

Perceived freshness and quality influence consumer purchases of beef (Brewer et al., 2002). Temperature affects these traits and the deterioration of food. Temperature affects not only the type and rate of bacterial growth (Giannuzzi et al., 1998), but also enzymatic spoilage (Bhattacharya et al., 2006). Despite the importance of ideal temperature, deviation from ideal temperature has been observed during storage and display of fresh beef (Koutsoumanis et al., 2006).

Modified-atmosphere packaging (MAP) containing oxygen promotes a cherry-red appearance. However, increased O₂ MAP increases lipid and pigment oxidation, resulting in off flavor, off odor, or dis-

coloration (Nawar, 1996). Furthermore, in MAP fresh beef held at high temperatures, the microbial and enzymatic spoilage reactions accelerate and the gas atmosphere can change (Limbo et al., 2010).

Synthetic antioxidants increase shelf life. However, consumers have indicated interest in natural products (Formanek et al., 2001) such as rosemary oleoresin (RO), which decreases microbial growth and metmyoglobin in packaged meat (Del Campo et al., 2000; Djenane et al., 2003; Camo et al., 2008).

Escherichia coli O157:H7 is an adulterant in raw ground beef, yet few food safety processing interventions exist. Previous research indicates that lactic acid bacteria (LAB) reduce pathogens such as *E. coli* O157:H7 and *Salmonella* spp. (Aguirre and Collins, 1993; Smith et al., 2005). Whereas prior work indicates benefits of LAB, some suggest that *Lactobacilli* (e.g., *Lactobacillus sake* L13; Egan et al., 1989) may have an effect on shelf life.

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Studies indicate that optimal shelf life is obtained at 0 to 4°C (Leak and Ronnow, 1999). However, few data exist regarding beef during abusive display. Also, few studies have examined the efficacy of interventions at abusive temperatures, defined by Limbo et al. (2010) as temperatures from 7 to 10°C. Therefore, the purpose of this study was to examine the effects of LAB and RO on the sensory traits of MAP ground beef displayed at abusive temperatures.

MATERIALS AND METHODS

Live animals were not used in this study; therefore, no approval from the Institutional Animal Care and Use Committee was obtained. Meat was obtained from a federally inspected meat processing facility.

Patty Preparation

A total of 104.5 kg of coarsely ground beef was obtained from a commercial beef-processing facility over a 3-wk period. A cocktail containing 4 strains of LAB (*Lactobacillus acidophilus* NP51, *Lactobacillus crispatus* NP35, *Pediococcus acidilactici*, and *Lactococcus lactis* ssp. *Lactis*) was provided by Culture Systems Inc. (Mishawaka, IN) for use at a targeted inoculation of 10⁹ cfu/g. The provided strains were preselected for antimicrobial activity, as illustrated by Amézquita and Brashears (2002) and Brashears et al. (2005). Before their addition to ground beef, the LAB strains were freeze dried and stored at -80°C. Inoculation levels were verified using the procedures outlined previously by Hoyle et al. (2009). For each replication (n = 3), ground beef was divided into 4 treatments: 1) control, without LAB or RO; 2) added RO (1,000 mg/kg) of RO added to fresh ground beef; 3) added LAB; and 4) added LAB and RO (LAB+RO). Control samples were prepared by mixing coarse-ground beef for 1 min, adding 250 mL of sterile distilled water (DW), and mixing for an additional 1 min using a commercial blender (model A-80, Koch Supplies Inc., Kansas City, MO). The RO- and LAB-treated ground beef was prepared in the same manner as the control, with the following exceptions: LAB was added in a 250-mL solution to provide 10⁹ cfu of LAB/g suspended in sterile DW, or RO (Herbalox Type HT-W, Kalsec Inc., Kalamazoo, MI) suspended in sterile DW was added at a level of 0.01% (1,000 mg/kg). Samples containing both LAB and RO were prepared by adding the previous concentrations of LAB or RO in 125 mL of sterile DW. After addition of RO or LAB or both, the coarse-ground product from each treatment group was finely ground using a 3.2-mm fine grind plate attached to a 3-phase meat grinder (model 346, Ciro, Ft. Smith, AR). Ground beef patties weighing approximately 145

g each were formed from each treatment group using a patty-forming machine (model 54, Hollymatic Corp., LaGrange, IL) and 2 patties were placed in each package.

Packaging

Rigid plastic trays measuring 27.3 cm × 17.21 cm × 7.6-cm deep (CS 978, Cryovac, Duncan, SC) were flushed with a gas mixture of 80% O₂/20% CO₂ and hermetically sealed using a film with oxygen transmission rates <20 cm³·m⁻²·24 h⁻¹ at 4.4°C and 100% relative humidity (LID 1050, Cryovac) using a gas-flush tray-sealing package machine (model CV/VG-S, G. Mondini S.p.a., Genoa, Italy). Packages were chosen randomly during packaging for analysis of gas composition using a headspace analyzer (CheckMate 9900, PBI Dansensor America, Glen Rock, NJ). All packages were visually inspected for leaks before retail display.

Simulated Retail Display and Temperature Abuse

Packages were displayed in a coffin-style retail display case (model M1, Hussman, Bridgeton, MO) maintained at 10°C. Temperature was monitored continuously using remote temperature recorders (Multi-Trip, Temprecord Monitor Co., Modesto, CA). Packages were subjected to an average of 1,900 lx of continuous fluorescent lighting using high-output bulbs (32 W) with a color temperature rating of 3,500°K and a color rendering index of 70. Packages were held in the retail cases for up to 36 h, with sensory analyses occurring every 12 h.

Trained Sensory Analysis and Consumer Evaluation

Both trained (n = 6 to 8) and consumer (n = 4 to 13) panelists were used to detect differences in color and odor among packaged patties at 12-h intervals during display. Panelists were trained by experienced meat science faculty in multiple sessions using representative samples before the start of the project. Trained panelists evaluated the lean color of ground beef patties using a 5-point, verbally anchored scale (1 = very bright red; 5 = very dark red or brown) as well as surface discoloration (1 = no discoloration; 5 = severe discoloration, 61 to 100%) according to color guidelines (AMSA, 1991). Nontrained graduate students were used as consumer panelists and were asked to determine whether the ground beef patties had good color (1 = very strongly agree; 7 = very strongly disagree) and how likely they were to purchase (1 = definitely would purchase; 5 = definitely would not purchase) the package based on the color (AMSA, 1991).

Odor panels were conducted on packages removed from the case at each sampling interval. The packages were opened in a random order and panelists were allowed to smell the patties without touching them. A verbally anchored numerical scale from Payne et al. (2002) was used for both trained and consumer panelists. Trained panelists were asked to determine whether an off odor was present (1 = no off odor; 5 = extreme off odor). Consumer panelists were asked whether the meat in the packaged smelled fresh (1 = very strongly agree; 7 = very strongly disagree) and how likely they were to consume the meat (1 = definitely would consume; 5 = definitely would not consume) based solely on the odor.

Objective Color Analysis

After sensory evaluation, CIE lightness (L^*), redness (a^*), and yellowness (b^*) values were taken from 1 patty per package. Color values were measured using a calibrated portable colorimeter (Hunter Miniscan XE Plus, model MSXP-4500C, Hunter Laboratories, Reston, VA) with illuminant D_{65} for CIE L^* , a^* , b^* and a standard observer angle of 10° and 2.54 cm aperture (CIE, 1978). Two observations were obtained from each patty and averaged to determine the respective CIE L^* , a^* , and b^* value. The CIE L^* , a^* , and b^* values were used to calculate hue angle ($\tan^{-1} b^*/a^*$) and saturation index $(a^{*2} + b^{*2})^{1/2}$.

Thiobarbituric Acid Reactive Substances Values

Thiobarbituric acid reactive substances (TBAR) values were analyzed as a measure of lipid oxidation using the procedures described by Luqué et al. (2011). Sample analyses were performed in duplicate for each patty after 0, 12, 24, and 36 h of display.

Statistical Analysis

The experimental design was a completely randomized split-plot design. Ground beef served as blocks to which treatment was assigned. Experiment was replicated 3 times. Statistical analyses were performed using the MIXED procedure (SAS Inst. Inc., Cary, NC) to evaluate the effect of LAB or RO treatment or a combination of both, display length (h), and any potential interaction on the trained and consumer sensory evaluations, instrumental color values, and lipid oxidation values of MAP packaged ground beef patties. Random variables included package identification, replication, and package identification \times replication. Significant main effects and interactions were analyzed using the least squares means method, and means were separated using the

PDIFF function of SAS. Differences were considered significant at $P < 0.05$ unless otherwise noted.

RESULTS AND DISCUSSION

Sensory Evaluation

Evaluations of trained panelists of lean color and discoloration did not differ between treatments ($P = 0.2273$ and 0.6592 , respectively; Table 1). Similarly, Camo et al. (2008) and Sawyer et al. (2009) found that rosemary extract had no effect on the lean color and discoloration of MAP lamb steaks stored for 11 or 7 d, respectively, at refrigeration temperatures. Rosemary oleoresin treatment affected the immediate off-odor scores from packaged ground beef patties, with RO and LAB+RO samples exhibiting reduced odor scores (less off odor) than ground beef patties without RO ($P < 0.0001$; Table 1). However, no effect on off odor was noted due to LAB inclusion ($P = 0.4894$). Similarly, Djenane et al. (2005) found no difference in off odors of MAP (70% O_2 :20% CO_2 :10% N_2) beef steaks when either *Lactobacillus sakei* or *Lactobacillus* CTC711 were applied to the steak surface and stored at $1^\circ C$.

An interaction was observed between treatment and display time ($P = 0.0940$; Table 2) for the lean color of MAP packaged ground beef stored at $10^\circ C$. Ground beef darkened as display progressed, regardless of treatment ($P < 0.0001$), but remained similar among all treatments throughout 24 h of display. However, after 36 h of display, samples containing RO had noticeably brighter lean color than samples without RO ($P < 0.05$). After 36 h of display, LAB ground beef was darker ($P < 0.05$) than the control ground beef. Previous research has noted darkening of lean during storage at $7^\circ C$ in beef steaks treated with

Table 1. Effect of lactic acid bacteria (LAB; 10^9 cfu/g) or rosemary oleoresin (RO; 1,000 mg/kg) or both on the evaluations by trained panelists of ground beef patties packaged in modified atmosphere and displayed at $10^\circ C$ for 36 h

Sensory trait	Treatment ¹				P-value	SEM
	Control	RO	LAB	LAB+RO		
Lean color ²	2.0	1.8	2.2	1.9	0.2273	0.16
Percent discoloration ³	1.3	1.2	1.3	1.2	0.6592	0.17
Immediate off odor ⁴	1.6 ^b	1.3 ^a	1.6 ^b	1.3 ^a	<0.0001	0.05

^{a,b}Least squares means within a row without a common superscript differ ($P < 0.05$).

¹Control = no addition of RO (1,000 mg/kg) or LAB (10^9 cfu/g); RO = addition of 1,000 mg/kg of RO only; LAB = addition of 10^9 cfu/g of LAB only; LAB+RO = addition of 1,000 mg/kg of RO and 10^9 cfu/g of LAB.

²1 = very bright red; 2 = bright red; 3 = slightly dark red or brown.

³1 = no discoloration; 2 = slight discoloration (1–10%).

⁴1 = no off odor; 2 = slight off odor.

Table 2. Effect of display (h) and treatment¹ (10⁹ cfu/g of lactic acid bacteria or 1,000 mg/kg of rosemary oleoresin) or both on the lean color, discoloration, and immediate off-odor scores by trained panelists of ground beef patties packaged in modified atmosphere and displayed at 10°C for 36 h

Sensory trait	Display, h				SEM
	0	12	24	36	
Lean color ²					0.21
Control	1.07 ^{a,z}	1.60 ^{b,z}	2.25 ^{c,z}	3.05 ^{d,y}	—
RO	1.16 ^{a,z}	1.50 ^{b,z}	2.11 ^{c,z}	2.57 ^{d,x}	—
LAB	1.23 ^{a,z}	1.54 ^{a,z}	2.43 ^{b,z}	3.47 ^{c,z}	—
LAB+RO	1.12 ^{a,z}	1.62 ^{b,z}	2.17 ^{c,z}	2.69 ^{d,xy}	—
Discoloration ³					0.12
Display main effects	1.00 ^a	1.02 ^a	1.22 ^a	2.04 ^b	—
Immediate off odor ⁴					0.12
Control	1.00 ^{a,z}	1.37 ^{b,y}	1.90 ^{c,y}	2.19 ^{d,x}	—
RO	1.00 ^{a,z}	1.11 ^{a,z}	1.47 ^{b,z}	1.44 ^{b,z}	—
LAB	1.00 ^{a,z}	1.25 ^{b,y}	1.90 ^{c,y}	2.11 ^{c,x}	—
LAB+RO	1.00 ^{a,z}	1.14 ^{a,yz}	1.41 ^{b,z}	1.63 ^{b,y}	—

^{a-d}Least squares means within a row lacking a common superscript differ ($P < 0.05$).

^{x-z}Least squares means within a column lacking a common superscript differ ($P < 0.05$).

¹Control = no addition of rosemary oleoresin (RO; 1,000 mg/kg) or lactic acid bacteria (LAB; 10⁹ cfu/g); RO = addition of 1,000 mg/kg of RO only; LAB = addition of 10⁹ cfu/g of LAB only; LAB+RO = addition of 1,000 mg/kg of RO and 10⁹ cfu/g of LAB.

²1 = very bright red; 2 = bright red; 3 = slightly dark red or brown; 4 = moderately dark red or brown. Treatment: $P = 0.2273$; display (h): $P < 0.0001$; treatment \times display (h): $P = 0.0940$.

³1 = no discoloration; 2 = slight discoloration (1–10%). Treatment: $P = 0.6592$; display (h): $P < 0.0001$; treatment \times display (h): $P = 0.3836$.

⁴1 = no off odor; 2 = slight off odor; 3 = small off odor. Treatment: $P < 0.001$; display (h): $P < 0.0001$; treatment \times display (h): $P = 0.0048$.

LAB; however, the LAB strains were different from those used in the present study (Leisner et al., 1995).

Similar to lean color scores, discoloration scores of trained panelists increased during the 36-h display period ($P < 0.0001$). No increase in discoloration was noted at 0, 12, and 24 h of display (1.00, 1.02, and 1.22, respectively; 1 = no discoloration). However, after 36 h of display, patties from all treatments exhibited slight discoloration (2.04; 2 = 1 to 19% discoloration). No differences were noted between LAB and the control ground beef at any display interval.

A treatment \times display length interaction was noted for the immediate off-odor scores of MAP ground beef patties ($P = 0.0048$; Table 2). No differences were noted among treatments at the beginning of display; however, after 12 and 24 h, samples containing RO (RO and LAB+RO) had less detectable off odor than samples lacking RO ($P < 0.05$). No increase in detectable off odor was noted between 24 and 36 h of display in ground beef treated with RO, LAB, or LAB+RO. However, the presence of off odors increased during the additional 12 h of display for

Table 3. Effect of display (h) and treatment¹ (10⁹ cfu/g of lactic acid bacteria or 1,000 mg/kg of rosemary oleoresin) or both on consumer panelist evaluations of ground beef patties packaged in modified atmosphere and displayed at 10°C for 36 h

Main effect	Sensory trait			
	Lean color ²	Purchase intent ³	Freshness of odor ⁴	Likelihood of consumption ⁵
Treatment				
Control	2.1 ^{y,z}	1.6	2.4 ^z	1.8
RO	1.9 ^y	1.5	2.1 ^x	1.6
LAB	2.2 ^z	1.7	2.4 ^{y,z}	1.7
LAB+RO	1.9 ^y	1.4	2.2 ^{xy}	1.6
<i>P</i> -value	0.0468	0.2951	0.0344	0.0649
SEM	0.13	0.13	0.10	0.08
Display, h				
0	1.53 ^z	1.17 ^z	1.55 ^z	1.11 ^z
12	1.50 ^z	1.13 ^z	1.78 ^z	1.43 ^y
24	2.13 ^y	1.57 ^y	2.58 ^y	1.83 ^x
36	2.99 ^x	2.55 ^x	3.11 ^x	2.31 ^w
<i>P</i> -value	<0.0001	<0.0001	<0.0001	<0.0001
SEM	0.15	0.10	0.13	0.08

^{w-z}Least squares means within a column and main effect lacking a common superscript differ ($P < 0.05$).

¹Control = no addition of rosemary oleoresin (RO; 1,000 mg/kg) or lactic acid bacteria (LAB; 10⁹ cfu/g); RO = addition of 1,000 mg/kg of RO only; LAB = addition of 10⁹ cfu/g of LAB only; LAB+RO = addition of 1,000 mg/kg of RO and 10⁹ cfu/g of LAB.

²“Do the patties have good color?” 1 = very strongly agree; 2 = strongly agree; 3 = slightly agree.

³1 = definitely would purchase; 2 = probably would purchase; 3 = may or may not purchase.

⁴“Does the meat in the package smell fresh?” 1 = very strongly agree; 2 = strongly agree; 3 = slightly agree; 4 = no opinion.

⁵1 = definitely would consume; 2 = probably would consume; 3 = may or may not consume.

control samples ($P < 0.05$). Regardless, at the conclusion of display, samples containing RO expressed the least off odor ($P < 0.05$). Similarly, Sánchez-Escalante et al. (2001) found less off odor in RO ground beef than controls over a 20-d display period, with noticeable differences occurring at d 4. These results indicate that the addition of RO may delay beef-quality deterioration; however, they do not indicate any effect or detriment attributable to LAB. Similar results were noted by Djenane et al. (2005).

No treatment \times display time interaction was noted for the evaluation of consumer panelists of lean color ($P = 0.2310$), purchase intent ($P = 0.1804$), freshness of odor ($P = 0.3450$), or likelihood of consumption ($P = 0.5981$; Table 3). However, treatment and display time (h) independently affect each trait. Consumers indicated ground beef patties lacking RO had less desirable color ($P < 0.05$; greater lean color scores) than patties with RO. No differences in lean color scores were noted between the control and LAB ground beef ($P = 0.6332$). Furthermore, consumer purchase intent scores indicated

no discernible difference in purchase intent between treatments ($P = 0.2951$; Table 3).

Modified-atmosphere packages with increased oxygen concentrations are associated with increased lipid oxidation (Zhao et al., 1994), resulting in a simultaneous increase in off odors and off flavors associated with lipid oxidation byproducts (Jakobsen and Bertelsen, 2000). The antioxidative effects of RO were noted in more favorable odor freshness scores for ground beef containing RO, in agreement with previous research by Brooks et al. (2008). Similarly to consumer evaluation of lean color, no differences in odor were detected due to LAB inclusion ($P = 0.9174$). Ground beef containing RO not only produced more favorable lean color and odor freshness, it also tended to increase consumers likelihood to consume the patties ($P = 0.0649$).

Length of display (h) affected the consumer sensory evaluations of MAP packaged ground beef patties (Table 3). Consumer lean color scores indicated less favorable lean color after 24 and 36 h of display (2.13 and 2.99, respectively; $P < 0.05$); however, scores did not change during the initial 12 h of retail display ($P = 0.8477$). Consumer purchase intent scores followed a similar trend and declined after 12 h of display ($P < 0.05$). These results coincide with previous research efforts that have documented the detrimental effects of display on meat color (Kropf, 1980; Brooks et al., 2008).

Odor freshness scores of MAP packaged ground beef patties increased (became less fresh; $P < 0.0001$) as display increased (Table 3). As with lean color and purchase intent scores, no difference was noted after 12 h of display ($P = 0.1491$), but increases were documented after 24 and 36 h (2.58 and 3.11, respectively; $P < 0.05$). Brooks et al. (2008) also noted an increase in consumer odor scores as display increased; however, samples in the study were not displayed at abusive temperatures. Vaikousi et al. (2009), who examined the effects of storage temperature on microbial growth and organoleptic properties of minced beef, indicated significantly stronger off-odor production in product exposed to 10°C when compared with storage temperatures of either 0 or 5°C.

Similarly to purchase intent scores, consumers became less likely to consume the product as display time increased ($P < 0.05$; Table 3). By 36 h, consumers indicated some probability to consume the packaged ground beef based solely on odor (2.31; 2 = probably would purchase).

Objective Color

The effects of RO and LAB addition to ground beef on instrumental color values are illustrated in Table 4. Hunter L^* values were not different among treatment groups ($P = 0.3110$). However, ground beef containing RO was more red (greater a^* values; $P < 0.05$) than samples without RO.

Table 4. Effect of lactic acid bacteria (LAB; 10^9 cfu/g) or rosemary oleoresin (RO; 1,000 mg/kg) or both on the instrumental color values of ground beef patties packaged in modified atmosphere and displayed at 10°C for 36 h

Color value ¹	Treatment ²				P-value	SEM
	Control	RO	LAB	LAB+RO		
L^*	48.59	48.43	48.47	48.84	0.3110	0.23
a^*	21.09 ^a	23.42 ^b	21.39 ^a	22.27 ^{ab}	0.0070	0.67
b^*	21.66 ^a	22.43 ^b	21.88 ^{ab}	22.45 ^b	0.0182	0.29
Hue angle ³	0.80 ^a	0.77 ^b	0.80 ^a	0.79 ^a	0.0138	0.01
Saturation ⁴	30.24 ^a	32.48 ^c	30.61 ^{ab}	31.63 ^{bc}	0.0042	0.61

^{a-c}Least squares means within a row lacking a common superscript differ ($P < 0.05$).

¹ L^* = lightness; a^* = redness; b^* = yellowness.

²Control = no addition of RO (1,000 mg/kg) or LAB (10^9 cfu/g); RO = addition of 1,000 mg/kg of RO only; LAB = addition of 10^9 cfu/g of LAB only; LAB+RO = addition of 1,000 mg/kg of RO and 10^9 cfu/g of LAB.

³Hue angle = $\tan^{-1} b^*/a^*$.

⁴Saturation = $(a^{*2} + b^{*2})^{1/2}$.

Similarly, Djenane et al. (2003) noted increased redness in ground beef containing RO. This is likely indicative of the antioxidative effect of RO on the conversion of oxy-myoglobin to metmyoglobin, as illustrated previously by Sánchez-Escalante et al. (2001). Furthermore, these results correspond with evaluations by consumer and trained panelists of lean color during display. Differences in Hunter b^* values were also noted among treatments ($P = 0.0182$), with control samples expressing the least values ($P < 0.05$), indicating increased metmyoglobin accumulation.

Hue angle and saturation index values of MAP packaged ground beef patties displayed at 10°C were affected by RO and LAB treatment ($P = 0.0138$ and 0.0042, respectively; Table 4). The increase in hue angle values because of greater temperatures has been documented (Limbo et al., 2010) and can be attributed to the increasing discoloration due to myoglobin oxidation (Mancini and Hunt, 2005). In the present study, the oxidation of myoglobin was deterred by the addition of RO, resulting in decreased hue angle values (less discoloration; $P < 0.05$). These results do not agree with trained panelist responses, which failed to indicate a difference in discoloration due to RO treatment (Table 1). However, the effect of RO treatment on the discoloration scores of trained panelists became apparent as display time increased.

The MAP ground beef patties containing RO exhibited a greater degree of red saturation ($P < 0.05$; greater saturation index; Table 4) than control patties. These results were in accordance with trained panelist evaluations, which were indicative of a brighter red lean color in samples containing RO. Samples containing only LAB had saturation values similar to those of either control or LAB+RO, suggesting the addition of LAB had no effect on the redness of ground beef.

TBAR Values

Treatment and display length interacted to affect TBAR values of MAP packaged ground beef patties ($P < 0.0001$; Table 5). No differences were observed among treatments at the beginning of display (0 h). However, after 12, 24, and 36 h of display, TBAR values from ground beef patties containing RO (RO and LAB+RO) were decreased ($P < 0.05$), indicating decreased lipid oxidation. Similarly, Ahn et al. (2007) and Sánchez-Escalante et al. (2001) noted no increase in lipid oxidation byproducts in ground beef samples containing RO.

High-oxygen MAP promotes oxidation and has been associated with increased TBAR values (O'Grady et al., 2000). Furthermore, Limbo et al. (2010) has documented an increase in TBAR values for ground beef held at 8 and 15.5°C when compared with 4.3°C. Therefore, an increase in TBAR values during display would be expected in the current study. The TBAR values from ground beef patties without RO increased as display time increased ($P < 0.05$; Table 5). However, no increase in TBAR values was noted in samples containing RO as display time increased from 0 to 36 h. Deterred lipid oxidation via the utilization of RO has been previously noted by Ho et al. (1995) and Barbut et al. (1985).

Green and Cumuze (1981) reported that 2 mg of malonaldehyde/kg of meat is required for the presence of rancid odors. In the current study, samples without RO expressed more than 2 mg of malonaldehyde/kg of sample after only 12 h of display. These results are in accordance with trained panelist evaluations, which were also indicative of an increase in off odor after only 12 h of display.

Table 5. Effect of display (h) and treatment (10^9 cfu/g of lactic acid bacteria or 1,000 mg/kg of rosemary oleoresin) or both on the thiobarbituric acid reactive substances (mg of malonaldehyde/kg of meat) for ground beef patties packaged in modified atmosphere and displayed at 10°C¹

Treatment ²	Display, h				SEM
	0	12	24	36	
Control	1.30 ^{a,z}	2.86 ^{b,z}	3.90 ^{c,z}	3.44 ^{bc,z}	0.44
RO	1.06 ^{a,z}	1.24 ^{a,x}	1.26 ^{a,y}	0.73 ^{a,y}	0.44
LAB	1.32 ^{a,z}	2.41 ^{b,y}	3.24 ^{c,z}	3.82 ^{c,z}	0.44
LAB+RO	1.27 ^{a,z}	1.22 ^{a,x}	1.20 ^{a,y}	0.68 ^{a,y}	0.44

^{a-c}Least squares means within a row lacking a common superscript differ ($P < 0.05$).

^{x-z}Least squares means within a column lacking a common superscript differ ($P < 0.05$).

¹Treatment × display (h): $P < 0.0001$.

²Control = no addition of rosemary oleoresin (RO; 1,000 mg/kg) or lactic acid bacteria (LAB; 10^9 cfu/g); RO = addition of 1,000 mg/kg of RO only; LAB = addition of 10^9 cfu/g of LAB only; LAB+RO = addition of 1,000 mg/kg of RO and 10^9 cfu/g of LAB.

Although spoilage is commonly thought to depend solely on the presence of microorganisms, it also depends on the numerous biochemical changes that occur in fresh meat. Mancini and Hunt (2005) state that color is an indicator of freshness and wholesomeness and is often a key factor in purchase decisions (O'Grady et al., 2000). Previous research in our laboratory has shown that the addition of this 4-strain LAB cocktail to ground beef inhibits the growth of *E. coli* O157:H7 and *Salmonella* spp. (Smith et al., 2005). Furthermore, Hoyle et al. (2009) found no difference in spoilage bacteria growth in ground beef patties treated with LAB or LAB+RO. Evidence regarding the general effects of LAB on microbial spoilage exists; however, consumers rely solely on perceived sensory characteristics, not microbial populations, to make purchase decisions of product in the retail case. For LAB to be properly used in the meat processing industry, a broader understanding of its effects (with or without RO) on the physicochemical and sensorial properties of ground beef shelf life is needed.

In conclusion, this research illustrates that the LAB strains used in this trial can be added to fresh ground beef without detrimentally affecting shelf life or the benefits afforded by addition of RO, which include prolonged color life and deterred lipid oxidation. These data, as supported by Hoyle et al. (2009), suggest that RO incorporation does not deter the growth of spoilage bacteria or LAB and enhances oxidative stability. The LAB strains used in this study do not grow significantly at refrigeration temperatures, but do produce compounds that inhibit the growth of pathogens such as *E. coli* O157:H7 and *Salmonella* spp. (Smith et al., 2005). Furthermore, microbial analyses (Hoyle et al., 2009) indicated no differences in spoilage microbial growth because of LAB treatment at abusive temperatures.

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