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Frozen storage stability of antioxidant-treated raw restructured beef steaks made from mature cows ☆

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Abstract

Previous research has shown that beef quality decreased with the age of cattle. In this study, beef trimmings from nine mature cows ($n = 9$), equally representing three animal age groups (2–4, 6–8, and 10–12 yr), were restructured into steaks formulated with propyl gallate, alone or in combination with a beefy flavoring agent, to enhance palatability and stability during 6 months of frozen storage at −29 °C. Lipid oxidation, rancidity, and loss of beefy flavor in restructured steaks during extended storage were reduced by propyl gallate. The beefy flavoring agent inclusion masked mature, forage-fed beef off-flavors, intensified beefy flavor, and improved steak tenderness, juiciness and cooking yield. Thus, the combination of propyl gallate and beefy flavoring offers an effective means to enhance the palatability and storage stability of restructured beef prepared from mature cows.

Keywords: Antioxidant; Flavor; Mature beef; Lipid oxidation; Restructured beef; Frozen storage

1. Introduction

The economic pressure on the meat industry to minimize production costs has served as incentives to develop new products of high quality and value from traditionally underutilized and/or less expensive fresh meat raw materials. Mature cattle, particularly dairy cows, have long been a vital contributor to the beef supply, representing 14.7% (head basis) of the total US beef slaughter in 2005 (USDA, 2007). However, limited merchandizing options exist for beef from advanced maturity, forage-fed cow carcasses due to inferior tenderness and flavor (Smith et al., 1982). It is well documented that palatability, sensory panel tenderness, connective tissue amount, and flavor scores deteriorated progressively as carcass maturity increased from C to E (Hilton et al., 1998). Our preliminary results confirmed that intact beef steaks from more mature cows (>10 yr of age) were significantly less tender than steaks from cows of younger ages (<4 yr) (data not shown).

A number of researchers have implicated prolonged forage consumption as a causative factor in the development of cooked beef off-flavors characterized as "grassy", "milky–oily", "soured dairy", "painty" and "fishy" (Hilton et al., 1998; Larick & Turner, 1990; Xiong, Moody, Blanchard, Liu, & Burris, 1996). It has also been documented that consumption of forages leads to increased concentrations of polyunsaturated fatty acids in beef muscles (Baublits et al., 2006; Larick & Turner, 1990; Marino et al., 2006; Srinivasan, Xiong, Blanchard, & Moody, 1998; Yang, Lanari, Brewster, & Tume, 2002), and an increase in susceptibility to oxidation and rancid off-flavor development during refrigerated or frozen storage. Meat restructuring technology can reduce the influence of connective tissue on final product tenderness and allows for the easy incorporation of ingredients that can potentially enhance storage stability and mask off-flavors inherent to mature beef (Schwartz & Mandigo, 1976).

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The objective of this study was to determine the effect of animal chronological age on restructured steak palatability and to test the hypothesis that antioxidant and beefy flavoring agents would reduce the objectionable flavors associated with mature, forage-fed cattle with a simultaneous minimization of lipid oxidation and rancidity development during extended period of frozen aerobic storage.

2. Materials and methods

2.1. Experimental design, animal production, and carcass handling

Nine cows ($n = 9$) composed predominantly of Angus × Simmental genetics were purchased from local producers with established record-keeping programs to ensure the genetic integrity and chronological age of each animal. Three different age categories were equally represented by the inclusion of three animals in each of the following age groups: (1) 2–4; (2) 6–8; and (3) 10–12 years of age. To establish more specific age intervals, all animals utilized were calved in the spring of the year. Cows selected as experimental units were acquired as they became available and placed on fescue pasture for a minimum of 2 months prior to slaughter.

Approximately 12 h prior to slaughter, cows were removed from both feed and water and transported to the USDA-inspected University of Kentucky abattoir where they were humanely slaughtered using standard industry procedures. After evisceration, carcasses were electrically stimulated with 70 1-s impulses, 550 V, 2.2 A, using a Boss electrical stunner (Model No. 1004D, Cincinnati Butcher Supply, Cincinnati, OH). After a 2 min rest period, carcasses were subjected to a series of 25 1-s impulses (Schaake, Means, Moody, Boyle, & Aaron, 1993). Carcasses were split immediately following electrical stimulation and chilled at 2 °C for 24 h, after which they were ribbed between the 12th and 13th ribs. Carcass maturity, based on both lean color and bone ossification on an A00 to E100 maturity scale to establish physiological age (USDA, 1996), was found to be C^{90}, D^{98}, and E^{93}, respectively, for the 2–4, 6–8, and 10–12 yr age groups.

2.2. Beef steak formulation and preparation

The chuck, plate, and brisket from both sides of each carcass were deboned and trimmed free of excessive fat and heavy connective tissue. The lean was packaged in 3 mil standard barrier nylon/PE vacuum pouches (Cryovac Sealed Air Corporation, Duncan, SC), chilled at 2 °C for 3 days, and then stored frozen at −29 °C for less than 3 months. Prior to restructuring, frozen lean meat in bags was thawed at 2 °C for 24 h and subsequently ground (Hobart Model 4116 SS grinder, Hobart Co., Troy, OH) through a 15.9 mm orifice plate. Ground meat samples were collected and immediately subjected to triplicate fat content determinations using a modified Babcock fat analysis test (Salwin, Bloch, & Mitchell, 1955). Lipid content of all ground meat batches was adjusted upward to a target level of 8% through the addition of frozen tallow (subcutaneous and intermuscular fat) obtained from the same carcass and previously ground through a 3.2 mm orifice plate.

After uniformly distributing the added fat throughout each batch by hand, 27.3 kg of lean from each carcass were equally divided into three 9.1-kg portions. Each portion was randomly assigned to one of the following three product formulation treatments: control (CTL) [1.5% NaCl and 0.25% sodium tripolyphosphate (STP)]; antioxidant (AO) [1.5% NaCl, 0.25% STP, and 0.02% propyl gallate on a fat content basis]; and antioxidant + beefy flavoring (AOBF) [1.5% NaCl, 0.25% STP, 0.02% propyl gallate, and 0.75% beefy flavoring (F&C Wild Flowers Inc., Cincinnati, OH)]. The restructuring process started with blending the mixed lean and fat for 2 min and then proceeded with the addition of ingredients using a Hobart upright mixer (Model A-200, Hobart Co., Troy, OH). Individual ingredients were sprinkled onto the meat during the mixing process in 2 min intervals in the following sequence: propyl gallate (to prevent salt-induced oxidation), STP, NaCl, and beefy flavoring.

Each batch was mixed for a total of 10 min independent of the number of ingredients incorporated. To provide uniform distribution, the propyl gallate was pre-dissolved in 10 ml of distilled water prior to addition. After mixing, each batch of formulated products was further divided into two equal portions (4.55 kg) and formed into restructured loaves using rectangular spring-loaded ham molds lined with polyvinyl chloride film. The compressed restructured loaves were stored at −29 °C for 1 h, removed from the molds, and allowed to freeze throughout at −29 °C for 24 h. Both frozen loaves per formulation were sliced with a mechanical band saw into ten 2.54-cm thick steaks to obtain a total of twenty restructured steaks per formulation per carcass.

2.3. Storage

Steaks were randomly distributed in the following manner: one steak for initial microbiological analysis; one steak, further divided into quarters, for raw lipid oxidation analysis at 0, 1, 3, and 6 month of frozen storage (−29 °C); four steaks for color, cooking yield, and textural evaluation at 0, 1, 3, and 6 months of frozen storage; and four steaks for sensory panel evaluation at the conclusion of each storage period. Samples for lipid oxidation analysis were stored without vacuum in 510 g capacity Whirl-Pak sterile bags (Fisher Scientific, Pittsburgh, PA). Steaks utilized for color, cooking yield, textural, and sensory panel evaluation were individually packaged without vacuum in wax coated meat freezer paper.

2.4. Microbiological evaluation

The initial microbial loads in restructured steaks (before storage) were examined using procedures of FDA (1998).

Specifically, three methods were utilized for identification and enumeration of microorganisms in samples: aerobic plate count agar incubated at 26 °C for 72 h to detect total psychrophiles; aerobic plate count agar incubated at 35 °C for 48 h to detect total mesophiles; and Violet Red Bile agar for total coliform count.

2.5. Measurement of lipid oxidation

Lipid oxidation was measured, in raw as well as cooked steaks, using the thiobarbituric acid (TBA) assay as described by Sinnhuber and Yu (1977). The TBA-reactive substances (TBARS), expressed as mg of malonaldehyde per kg of meat, was calculated as TBARS (mg/kg) = $(A_{520}/W_s) \times 9.48$, where W_s is the meat sample weight and the value 9.48 is a constant derived from the sample dilution and the absorption coefficient (152,000 M^{-1} cm^{-1}) of the TBA-malonaldehyde adduct.

2.6. Color evaluation

A HunterLab Digital Color Difference Meter (Model D2D4 M/LL, Fairfax, VA) was used to determine L (luminosity), a (redness), and b (yellowness) values, saturation index $((a^2 + b^2)^{1/2})$, and hue angle $(\tan^{-1} b/a)$ of thawed (2 °C for 16 h) beef steaks according to AMSA (1991). The instrument was calibrated with HunterLab standard plate No. C2-13717, with $L = 68.6$, $a = 23.5$, and $b = 12.8$. Three measurements were taken on each steak at different sites uniformly spaced over the surface and the colorimetric values were averaged.

2.7. Cooking

Thawed (at 2 °C for 16 h) steaks were cooked to an internal temperature of 70 °C on a Farberware Open Hearth electric broiler (Farberware Inc., Bronx, NY). The center temperature was monitored with a handheld thermocouple (Model 31308-KF, Type K thermocouple, Atkins Technical Inc., Gainesville, FL). Cooking yield was defined as the cooked meat sample weight (g) divided by raw meat weight (g) then multiplying by 100. Cooked steaks were used immediately for subsequent textural analysis.

2.8. Textural measurements

An Instron Universal Testing Machine (Model 4301, Instron Corp., Canton, MA) was used to determine rigidity and rupture force of cooked restructured beef steaks as previously described by Xiong, Noel, and Moody (1999). Samples were prepared with a razor sharp knife by cutting steaks into cubes measuring 15 mm^3 (with the cooked surfaces removed). The cubes were placed between two parallel plates and compressed at a 200 mm/min crosshead speed to obtain both a 20% and a 80% sample height reduction. The compressive force generated by a 20% height reduction was used to indicate sample hardness (rigidity). An 80% sample deformation resulted in structural failure of all samples. The point of structural failure was identified by a sharp rise and fall in compressive force and was used to determine the rupture force (binding strength). Ten samples were prepared and compressed for each steak and the mean values calculated for hardness and binding strength.

2.9. Sensory evaluation

Cooked steak palatability characteristics were evaluated by an eight-member trained panel in a sensory evaluation laboratory with partitioned booths illuminated by red lights to mask color differences between samples. The panelists were selected faculty, staff, and graduate students who had previously participated in meat sensory evaluation and trained in three training sessions according to AMSA (1995). Nine samples were evaluated per sensory panel session with a maximum of one session per day and two sessions per week. Samples were provided to the panelists in a random fashion during each session. Panelists evaluated steak samples for tenderness, juiciness, beefy flavor, off-flavor, rancidity, and overall acceptability on a numeric scale of 1–8 (1 = extremely tough, dry, bland, unacceptable; 2 = very tough, dry, bland, unacceptable; 3 = moderately tough, dry, bland, unacceptable; 4 = slightly tough, dry, bland, unacceptable; 5 = slightly tender, juicy, intense, acceptable; 6 = moderately tender, juicy, intense, acceptable; 7 = very tender, juicy, intense, or acceptable; and 8 = extremely tender, juicy, intense, acceptable).

2.10. Statistical analysis

The experiment was a randomized complete block design, using slaughter group to establish blocks, namely, one cow from each maturity group was slaughtered on the same day. All data were analyzed using the GLM procedures of SAS (SAS Inst. Inc., Cary, NC). Data were analyzed as a split-plot with animal age as the whole-plot factor, product formulation as the sub-plot factor and storage time as the sub-sub-plot factor. All main effect and interaction means were separated using least square procedures when the respective F-tests were found significant ($P < 0.05$).

3. Results and discussion

3.1. Microbiological quality

Because good sanitation practices were followed, the initial level of contamination (aerobic count) was relatively low (3.18 and 3.05 $\log_{10}$ CFU/g for psychrotrophic and mesophilic counts, respectively) for all samples (data not shown). Coliforms were not detected in any of the samples.

3.2. Lipid oxidation

Lipid oxidation (TBARS) was not influenced by animal age but increased significantly ($P < 0.01$) in the absence of

propyl gallate, and over extended periods of storage (Table 1). Forage consumption was found to increase muscle polyunsaturated fatty acid concentration (Baublits et al., 2006; Larick & Turner, 1990; Marino et al., 2006; Srinivasan et al., 1998; Yang et al., 2002). As a result, beef from forage-fed cattle was more susceptible to lipid oxidation (Reverte, Xiong, & Moody, 2003; Xiong et al., 1996) than beef from grain-fed cattle. The mature cows used in the present study had similar nutritional background (i.e. forage-based), and as a function of chronological age, differed only in the length of time spent consuming forages. The lack of differences between animal age groups only suggested that when nutritional background is held constant, animal chronological age is of little consequence in determining lipid oxidative stability in muscle if ground and then restructured into steaks.

The specific comparison of steak formulations across storage time indicated no formulation influence on lipid oxidation for the first 3 months of storage (Table 2). With the exception of a marginal increase ($P < 0.05$) among CTL steaks, values did not change within formulations during this time. However, cooking slightly increased the TBARS content for all samples. In contrast, TBARS values increased drastically ($P < 0.05$) for CTL steaks over the last 3 months of storage (e.g., 3 months = 0.86 mg/kg and 6 months = 3.10 mg/kg for raw steaks), while values of AO and AOBF steaks were similar ($P > 0.05$) to each other and only slightly higher ($P < 0.05$) compared to earlier values.

These observations demonstrated that lipid oxidation increased as frozen storage progressed, but was effectively retarded when propyl gallate was incorporated into restructured beef steaks. The antioxidant effect explained the significant formulation × storage time interaction ($P < 0.05$) in these restructured meat samples. Our observations are in agreement with the earlier findings that TBARS values of frozen restructured beef increased over extended periods (Akamittath, Brekke, & Schanus, 1990; Crackel, Gray, Booren, Pearson, & Buckley, 1988; Lamkey, Mandigo, & Calkins, 1986; Reverte et al., 2003). In addition, Reverte et al. (2003) observed a similar antioxidant effect of propyl gallate in beef from young cattle.

Synthetic phenolic antioxidants as free radical scavengers have been shown to control lipid oxidation reactions in foods (Chastain, Huffman, Hsieh, & Cordray, 1982; Crackel et al., 1988). Phosphate was claimed to exert antioxidant effects due to its ability to chelate prooxidative metal ions (Arganosa, Godber, Tanchotikul, McMillin, & Shao, 1991; Lamkey et al., 1986). In the present study, the addition of 0.25% tripolyphosphate in CTL samples was apparently incapable of inhibiting lipid oxidation after 3 months of storage. Akamittath et al. (1990) reported that the prooxidative effect of salt and other substances, such as activated metmyoglobin-peroxide present in beef muscle, overcame the protective effect of the tripolyphosphate in steaks over extended storage periods. The findings from the current study suggested that restructured steaks formulated exclusively with beef from mature cows and stored frozen for more than 3 months should contain extraneous antioxidants, beyond the addition of tripolyphosphate, to minimize lipid oxidation and counteract the prooxidant effect of added salt. Steaks treated with propyl gallate displayed only minor lipid oxidation, suggesting that free radical chain reactions, rather than metal ion-dependent catalysis, may have been the main mechanism involved in oxidation of restructured beef during prolonged frozen storage (Reverte et al., 2003).

3.3. Color stability

Animal age did not influence ($P > 0.10$) any instrumental colorimetric reading of stored raw steaks (data not shown). Beef muscle color was reported to become darker with advancements in animal age (Powell, 1991; Tuma, Henrickson, Odell, & Stephens, 1963). It was likely that in the present study, either the maturity in all animal age groups had advanced enough to not reveal any color differences or the restructuring and sample preparation process (i.e. grinding and mixing, salt addition, and freezing and thawing) had diminished the age-dependent color difference between steaks (Chu, Huffman, Trout, & Egbert, 1987). It was noteworthy that steaks from the present study tended to be darker (lower L value) and redder (higher a value) than those found by Reverte et al. (2003) on beef from young maturity carcasses. This was in line with Powell (1991) who reported that the color of beef from younger animals was lighter and less red than that from older animals. As a result, steaks manufactured solely with beef from mature cows may not be optimally suited for raw steak merchandizing methods due to inherent color defects.

Because of the lack of animal age effect, the colorimetric values from all age groups were pooled by storage time for

Table 1
Main effect means of degree of lipid oxidation (TBARS, mg/kg) in raw restructured beef steaks stored −29 °C

Main effect												
Animal age (yr)				Formulation[d]				Storage time (month)				
2–4	6–8	10–12	SE	CTL	AO	AOBF	SE	0	1	3	6	SE
1.20	1.13	1.19	0.15	1.54[a]	1.03[b]	1.09[b]	0.04	0.86[c]	0.82[c]	1.15[b]	2.08[a]	0.05

[a–c] Wherever marked by a letter(s), mean values for the same main effect without a common letter differ significantly ($P < 0.01$).
[d] CTL = control; AO = antioxidant; AOBF = antioxidant + beefy flavoring; SE = standard error.

Table 2
Effect of formulation treatments and frozen (−29 °C) storage time on lipid oxidation (TBARS, mg/kg) in restructured beef steaks before (raw) and after cooking

Formulation[h]	Storage time (month)							
	0		1		3		6	
	Raw	Cooked	Raw	Cooked	Raw	Cooked	Raw	Cooked
CTL	0.52^{g}	1.24^{cd}	0.62^{fg}	0.95^{de}	0.86^{ef}	1.55^{c}	3.10$^{b/x}$	3.58$^{a/x}$
AO	0.58^{d}	1.01^{b}	0.68^{cd}	0.99^{bc}	0.80^{bcd}	1.49^{a}	0.97$^{bc/y}$	1.68$^{a/y}$
AOBF	0.74^{d}	1.14^{bc}	0.74^{d}	0.95^{cd}	0.95^{cd}	1.28^{b}	1.17$^{bc/y}$	1.72$^{a/y}$

Standard error = 0.55 (formulation × storage time × cooking interaction).
[a–g] Mean values in the same row without a common letter differ significantly ($P < 0.05$).
[h] CTL = control; AO = antioxidant; AOBF = antioxidant + beefy flavoring.
[x,y] Wherever marked by a letter(s), mean values in the same column without a common letter differ significantly ($P < 0.05$).

analysis. As displayed in Table 3, the raw steak color (*a*, and *b* values and saturation indexes) deteriorated progressively from month 0 to month 6 ($P < 0.01$). These colorimetric values as well as their changes during storage were similar ($P > 0.05$) for CTL and AO raw steaks. The slightly lower *a* and saturation index values for AOBF steaks were attributed to the incorporated beef flavoring that was dark brown in color. In agreement with the findings of Reverte et al. (2003), these data suggested that propyl gallate was ineffective in retarding heme pigment oxidation (i.e. conversion of ferrous ion to ferric ion in the heme complex) and supported the findings that color attributes of restructured beef steaks (Schaake et al., 1993) and formed pork and turkey products (Akamittath et al., 1990) decreased during aerobic storage. Huffman, McCafferty, Cordray, and Stanley (1984) stated that addition of salt to restructured beef products increased discoloration of raw steaks, and suggested that the added salt might act as a prooxidant by interacting with the heme and reducing the pH of the meat product. An alteration of the ionic environment of the heme cleft in the myoglobin molecule may have caused a destabilization of the heme structure, allowing oxidation of the heme iron to occur (Fox, 1966). As shown by Seideman, Cross, Smith, and Durland (1984), salt can also act as a prooxidant and promote pigment oxidation by reducing the oxygen tension and decreasing the buffering capacity of meat, thereby increasing the potential for myoglobin oxidation.

Table 3
Effect of formulation treatments and frozen (−29 °C) storage time on instrumental colorimetric values of raw restructured beef steaks

Treatment color parameter[e]	Storage time (month)			
	0	1	3	6
L value (SE = 0.23)				
CTL	27.7	25.5	25.9	25.8
AO	27.3	25.7	26.2	25.7
AOBF	26.9	25.7	26.5	25.8
a value (SE = 0.25)				
CTL	26.4$^{a/x}$	25.2$^{b/x}$	22.1$^{c/x}$	19.1^{d}
AO	26.2$^{a/x}$	24.3$^{b/y}$	21.7$^{c/x}$	19.6^{d}
AOBF	23.2$^{a/y}$	21.5$^{b/z}$	19.2$^{c/y}$	19.2^{c}
b value (SE = 0.10)				
CTL	10.3$^{a/x}$	29.2$^{b/x}$	28.3^{c}	28.1^{c}
AO	10.1$^{a/x}$	29.1$^{b/x}$	28.1^{c}	28.1^{c}
AOBF	19.4$^{a/y}$	28.7$^{b/y}$	28.2^{c}	28.0^{c}
Saturation index (SE = 0.25)				
CTL	28.4$^{a/x}$	26.8$^{b/x}$	23.6$^{c/x}$	20.7^{d}
AO	28.1$^{a/x}$	26.0$^{b/y}$	23.2$^{c/x}$	21.1^{d}
AOBF	25.0$^{a/y}$	23.2$^{b/z}$	21.0$^{c/y}$	20.8^{c}
Hue angle (SE = 0.25)				
CTL	21.3$^{b/y}$	20.2$^{c/y}$	20.6$^{c/y}$	23.2$^{a/x}$
AO	21.2$^{ab/y}$	20.5$^{b/y}$	20.5$^{b/y}$	21.7$^{a/y}$
AOBF	22.1$^{b/x}$	22.0$^{b/x}$	23.1$^{a/x}$	22.7$^{ab/x}$

[a–d] Wherever marked by a letter(s), mean values in the same row without a common letter differ significantly ($P < 0.05$).
[e] CTL = control; AO = antioxidant; AOBF = antioxidant + beefy flavoring; SE = standard error.
[x–z] Wherever marked by a letter(s), mean values for the same color parameter in the same column without a common letter differ significantly ($P < 0.05$).

The inclusion of a beefy flavoring agent decreased steak *a* values, *b* values, and saturation index at 0 month compared to CTL and AO steaks (Table 3). Interestingly, these colorimetric values of AOBF steaks remained lower ($P < 0.01$) than the values of CTL and AO steaks for 1 or 3 months, but were similar ($P > 0.05$) across formulations by 6 months when the latter samples experienced a significant value drop in these colorimetric parameters. These observations suggested that the low initial colorimetric values in AOBF steaks were caused by the pigments from the beefy flavoring agent that effectively masked the raw beef color of the steaks, and were not a result of myoglobin oxidation. The presence of the flavoring agent seemed to retard muscle pigment oxidation during extended frozen storage.

3.4. Cooking yield

Animal age did not influence ($P > 0.10$) cooking yield; however, significant ($P < 0.01$) formulation and storage time main effects were revealed (Table 4). Inclusion of a beefy flavoring agent increased ($P < 0.01$) cooking yield, while propyl gallate alone had no effect ($P > 0.10$). Other researchers reported similar findings, suggesting that the yeast extract incorporated as a component of the beefy flavoring agent (up to 47% of the beefy flavoring agent) may

contribute to water-binding in cooked steaks (DeYonge-Freeman, Pringle, Reynolds, & Williams, 2000; Reverte et al., 2003; Scanga et al., 2000). Cooking yield did not change during the first month of storage, but decreased ($P < 0.01$) after 3 and 6 months. Chastain et al. (1982) also showed that cooking yields of beef–pork (50:50) restructured steaks decreased as frozen storage progressed over 20 weeks, and implicated the physical disruption of cell structure or possible protein denaturation during frozen storage. In contrast, Reverte et al. (2003) reported that restructured steak cooking yield did not change during 6 months of frozen storage.

3.5. Textural measurements

Hardness of restructured steaks was not influenced ($P > 0.10$) by either animal age or formulation, although mean values tended to decrease slightly as storage time progressed beyond 1 month (Table 4). Similarly, no significant ($P > 0.10$) main effects or their interactions existed for restructured steak sample rupture force (Table 4), suggesting that all the steaks were equally well bound. The lack of differences in the measured textural properties between animal ages, product formulations, or storage times indicated the feasibility of restructuring as a means to diminish the variation in tenderness of beef from mature carcasses that have been widely reported in the literature (Hilton et al., 1998; Smith et al., 1982) and observed in our preliminary study. Consistent with these findings, Reverte et al. (2003) showed that binding strength of restructured beef steaks did not change during frozen storage and was not influenced by the inclusion of an antioxidant and/or beefy flavoring agent. Ockerman and Organisciak (1979) also noted that fresh restructured steak cohesiveness was not influenced by frozen storage up to 90 days. The results of the current study seemed to indicate that protein exudates, extracted by mixing of raw meat with salt and phosphate during the meat restructuring process, were not significantly denatured during frozen storage (Reverte et al., 2003). This conclusion was supported by the observations of Coon, Calkins, and Mandigo (1983) who reported that restructuring with salt and phosphates produced well-bound steaks and eliminated variation of the low quality beef cuts, thereby creating a uniform value-added product.

3.6. Sensory evaluation

While animal age did not influence ($P > 0.10$) most sensory traits (Table 5), the 10–12 yr age group steaks received a lower ($P < 0.05$) beefy flavor score than the other two age group steaks. Hilton et al. (1998) demonstrated that beefy flavor tended to become blander as carcass maturity increased, and Smith et al. (1982) reported that flavor acceptability decreased as carcass maturity progressed from C to E. The observed similarity ($P > 0.10$) in rancidity scores across animal age groups was supported by TBARS analysis (Table 1). Panelists rated the steaks as "slightly tender" and detected no difference ($P > 0.10$) between animal age groups, confirming that restructuring effectively eliminated the animal age-associated tenderness variation that was known to exist between groups.

As expected, propyl gallate did not influence ($P > 0.10$) steak tenderness or juiciness, but the inclusion of the beefy flavoring agent improved ($P < 0.01$) both traits, which was consistent with the cooking yield results (Table 4). The yeast extract contained in the flavoring agent probably contributed to water-binding and hence, a higher juiciness score. DeYonge-Freeman et al. (2000) and Scanga et al. (2000) arrived at a similar conclusion after improvements were observed in intact steaks injected or marinated with a flavoring agent. Of the three product formulations, AOBF steaks received the highest beefy flavor score ($P < 0.05$). The AO sample also had a more intense ($P < 0.05$) beefy flavor than control sample, probably due to its reduced rancidity that seemed to mask desirable flavors.

Storage time affected sensory attributes of restructured beef. Tenderness and juiciness scores decreased ($P < 0.01$) after 6 months. This again corresponded with a significant decrease ($P < 0.05$) in cooking yield. Toward the end of frozen storage (6 months), there were significant losses in beefy flavor while off-flavors and rancidity became more intense ($P < 0.01$).

Table 6 presents the mean panel scores of selected sensory attributes of the three steak formulations over their respective storage times. In agreement with a previous finding (Reverte et al., 2003), propyl gallate did not prevent the development of off-flavors inherent to beef from mature forage-fed cattle. Panelists characterized the off-flavors as "grassy", "metallic" and/or "livery". These descriptions

Table 4
Main effect means of cooking yield and instrumental textural properties of restructured steaks stored at −29 °C

Property	Main effect												
	Animal age (yr)				Formulation[d]				Storage time (month)				
	2–4	6–8	10–12	SE	CTL	AO	AOBF	SE	0	1	3	6	SE
Cooking yield (%)	83.6	84.7	85.6	0.51	83.7[b]	83.9[b]	86.2[a]	0.29	86.2[a]	85.5[ab]	84.7[b]	82.1[c]	0.42
Hardness (N)	15.0	14.1	14.4	0.44	14.4	14.4	14.7	0.35	14.8[ab]	15.4[a]	14.0[bc]	13.7[c]	0.37
Rupture force (N)	96.1	87.4	88.6	3.05	91.4	92.0	88.7	2.49	89.6	89.6	89.0	94.7	2.13

[a–c] Wherever marked by a letter(s), mean values for the same main effect in the same row without a common letter differ significantly ($P < 0.05$).
[d] CTL = control; AO = antioxidant; AOBF = antioxidant + beefy flavoring; SE = standard error.

Table 5
Main effect means of sensory panel scores of restructured steaks stored at −29 °C

Sensory attribute	Main effect												
	Animal age (yr)				Formulation[e]				Storage time (month)				
	2–4	6–8	10–12	SE	CTL	AO	AOBF	SE	0	1	3	6	SE
Tenderness	5.3	5.4	5.2	0.14	5.1^{b}	5.2^{b}	5.5^{a}	0.08	5.4^{a}	5.4^{a}	5.3^{a}	5.0^{b}	0.07
Juiciness	5.4	5.5	5.4	0.06	5.2^{b}	5.3^{b}	5.8^{a}	0.06	5.6^{a}	5.6^{a}	5.4^{a}	5.1^{b}	0.10
Beefy flavor	5.1^{a}	5.0^{a}	4.7^{b}	0.05	3.9^{c}	4.3^{b}	6.6^{a}	0.09	5.2^{a}	4.9^{ab}	5.2^{a}	4.5^{b}	0.13
Off-flavor	2.0	2.1	2.4	0.17	2.5^{a}	2.4^{a}	1.7^{b}	0.14	1.5^{c}	1.9^{b}	2.7^{a}	2.6^{a}	0.11
Rancidity	1.6	1.5	1.6	0.05	2.0^{a}	1.6^{b}	1.1^{c}	0.06	1.0^{c}	1.2^{bc}	1.4^{b}	2.7^{a}	0.09
Overall acceptability	4.9	5.0	4.7	0.08	4.4^{b}	4.6^{b}	5.7^{a}	0.11	5.5^{a}	5.2^{b}	4.8^{c}	4.0^{d}	0.10

[a–d] Wherever marked by a letter(s), mean scores for the same main effect in the same row without a common letter differ significantly ($P < 0.05$).
[e] CTL = control; AO = antioxidant; AOBF = antioxidant + beefy flavoring; SE = standard error. Score range: 1–8 (see Section 2).

Table 6
Effect of formulation treatments and frozen (−29 °C) storage time on the sensory panel scores of restructured steaks

Formulation sensory attribute[e]	Storage time (month)			
	0	1	3	6
Off-flavor (SE = 0.20)				
CTL	1.4^{d}	$2.2^{c/x}$	$3.4^{a/x}$	$2.8^{b/x}$
AO	1.7^{b}	$2.0^{b/xy}$	$2.9^{a/x}$	$2.8^{a/x}$
AOBF	1.4^{b}	$1.4^{b/y}$	$1.7^{ab/y}$	$2.2^{a/y}$
Rancidity (SE = 0.16)				
CTL	1.0^{c}	1.2^{c}	$1.8^{b/x}$	$4.1^{a/x}$
AO	1.0^{b}	1.3^{b}	$1.5^{b/xy}$	$2.5^{a/y}$
AOBF	1.0	1.0	1.0^{y}	1.5^{z}
Overall acceptability (SE = 0.18)				
CTL	$5.4^{a/y}$	$4.8^{b/y}$	$4.1^{c/y}$	$3.2^{d/y}$
AO	$5.2^{a/y}$	$4.8^{ab/y}$	$4.5^{b/y}$	$3.8^{c/y}$
AOBF	$5.9^{a/x}$	$5.9^{a/x}$	$5.8^{a/x}$	$5.1^{b/x}$

[a–d] Wherever marked by a letter(s), mean scores in the same row without a common letter differ significantly ($P < 0.05$).
[e] CTL = control; AO = antioxidant; AOBF = antioxidant + beefy flavoring; score range: 1–8 (See Section 2). SE = standard error.
[x–z] Wherever marked by a letter(s), mean scores for the same sensory trait in the same column without a common letter differ significantly ($P < 0.05$).

echoed those identified by other researchers that undesirable flavors of beef from forage-fed cattle were noted as "dairy", "fishy", "grassy", "milky", "oily", "painty" and/or "sour" (Davis, Cole, Backus, & Melton, 1981; Hilton et al., 1998; Mandell, Buchanan-Smith, & Campbell, 1998). In contrast, the addition of a beefy flavoring agent resulted in a notable reduction ($P < 0.05$) in detectable off-flavors and was effective in masking the undesirable "grassy" flavor throughout storage. DeYonge-Freeman et al. (2000) and Reverte et al. (2003) also reported similar observations.

Although rancidity scores increased ($P < 0.05$), notably beyond 3 months, propyl gallate delayed its onset (Table 6). The result was supported by TBARS values, which suggested that propyl gallate effectively retarded lipid oxidation. Rancidity was non-detectable for the first 3 months in AOBF steaks and was only slightly detected after 6 months. While TBARS values were similar for AOBF and AO steaks (Table 2), both rancidity and off-flavors were found to be less intense ($P < 0.01$) in AOBF samples than in AO samples at 6 months. Whether this was due to flavor-masking by the beefy flavoring agent or its ability to actually suppress the production of off-flavor compounds needs to be further researched. It appeared that the protection of desirable beefy flavor and at the same time, inhibition of off-flavor compounds, led the panelists to conclude that AOBF steaks were not only well acceptable but significantly more so than CTL and AO steaks ($P < 0.01$). The inclusion of a beefy flavoring agent may be an essential ingredient when steaks were manufactured with beef from mature cows and then stored frozen for up to 6 months.

4. Conclusions

The physiological age of mature cows had little influence on the palatability and frozen storage stability of restructured steaks. Thus, beef originating from mature cows can be processed into a homogeneous meat block in the form of restructured steaks. Propyl gallate successfully retarded lipid oxidation and the development of rancid flavors, but was unable to overcome the low acceptability of steaks with inherent off-flavors. However, when the antioxidant was used in combination with a beefy flavoring agent, a marked improvement in all sensory panel palatability traits was observed. Improvements in tenderness and juiciness were also associated with the inclusion of the beefy flavoring agent that reduced the cooking loss. Thus, incorporation of proper antioxidant(s) and flavoring agent(s) seem to be required for the manufacture of acceptable beef steaks from mature cows when subjected to frozen storage.

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