

# EFFECTS OF HOT-WATER EXTRACTS FROM MYRTLE, ROSEMARY, NETTLE AND LEMON BALM LEAVES ON LIPID OXIDATION AND COLOR OF BEEF PATTIES DURING FROZEN STORAGE

A. AKARPAT, S. TURHAN<sup>1</sup> and N.S. USTUN

*Department of Food Engineering  
Ondokuz Mayıs University  
55139 Samsun, Turkey*

Accepted for Publication August 22, 2007

## ABSTRACT

*The effects of hot-water extracts from myrtle, rosemary, nettle and lemon balm leaves on lipid oxidation and the color properties of beef patties during frozen storage were studied. Ground beef was treated with salt (1.5%) and five types of samples were prepared: control (no extract), 10% myrtle extract added, 10% rosemary extract added, 10% nettle extract added and 10% lemon balm extract added. Each group was adjusted to a fat content of 20% by the addition of backfat. Patties (25 g) were wrapped with polyethylene film and stored at  $-20 \pm 2^\circ\text{C}$  for 120 days. Oxidative and sensory changes were evaluated after 0, 30, 60, 90 and 120 days of storage. Hot-water extracts from myrtle, rosemary, nettle and lemon balm leaves affected peroxide value (POV), thiobarbituric acid-reactive substances (TBARS), color (except L value) and sensory properties of beef patties. Hot-water extracts from myrtle, rosemary, nettle and lemon balm leaves slowed down the lipid oxidation of beef patties. While myrtle and rosemary extracts showed the highest antioxidant effects, the effects of nettle and lemon balm extracts were lower. Myrtle extract protected the color properties of frozen beef patties. In terms of overall acceptability, myrtle and rosemary extracts were given the highest points by the panelists. Storage time affected pH, POV, TBARS, color (except L and b values) and sensory properties of beef patties. The addition of hot-water extracts from myrtle leaves at 10% ratio to the frozen storage of beef patties prevents development of oxidative processes in lipids and changes in color.*

<sup>1</sup> Corresponding author. TEL: +90-362-3121919; FAX: +90-362-4576094; EMAIL: sturhan@omu.edu.tr

## PRACTICAL APPLICATIONS

Lipid oxidation is one of the primary causes of quality losses in frozen stored meat and meat products. One of the methods for protection against oxidation is to use antioxidants. Hot-water extracts from myrtle and rosemary slowed down the lipid oxidation and prevented color changes of beef patties. According to these results, hot-water extracts of myrtle and rosemary can be used to extend the shelf life of meat patties and other meat products.

## INTRODUCTION

Freezing is an excellent process for preserving the quality of meat and fish for long periods. Its effectiveness stems from internal dehydration (ice crystal formation) or immobilization of water and lowering of temperature. Although some microorganisms survive storage at very low temperatures, there generally is no opportunity for growth of microorganisms if recommended storage temperatures are maintained (Foegeding *et al.* 1996). However, deterioration of quality caused by chemical or physical causes can occur. Studies have shown that lipid oxidation is one of the primary causes of quality losses in frozen stored meat and meat products (Gokalp *et al.* 1983). The oxidation process reduces the nutritional value of lipids greatly. The color alters as the pigments oxidize and the flavor and aroma also change as a result of the accumulation of secondary volatiles. Biologically active compounds can be destroyed and, in some cases, toxic and carcinogenic substances (represented by hydroperoxides, radicals, epoxides, aldehydes, acids, etc.) accumulate (Balev *et al.* 2005).

The retardation of the oxidation process is important for the food processor and, indeed, for all persons involved in the entire food chain from the factory to the consumer. Oxidation may be inhibited by various methods including prevention of oxygen access, use of lower temperature, inactivation of enzymes catalyzing oxidation, reduction of oxygen pressure and the use of suitable packaging. Another method of protection against oxidation is to use antioxidants (Pokorny 2001). Food antioxidants are compounds or substances that are able to inhibit or interfere with the free-radical autoxidation reaction fundamental to glyceride oxidation. This ability arises from their phenolic structure or a phenolic configuration within their molecular structure (Sherwin 1990). Literally hundreds of compounds, both natural and synthesized, have been reported to possess antioxidant properties. Their use in food, however, is limited by certain obvious requirements not the least of which is adequate proof of safety (Nawar 1996). In industrial processing, mainly synthetic antioxidants such as butylated hydroxyanisole, butylated hydroxytoluene and

propyl gallate are used, in order to prolong the storage stability of food. However, the demand for natural antioxidants has recently increased because of the toxicity and carcinogenicity of synthetic antioxidants (Juntachote *et al.* 2006). In addition, synthetic antioxidants have limited applications because of their low water solubility (Bekhit *et al.* 2003). Consequently, consumers generally perceive natural antioxidants as better than synthetic antioxidants.

Natural antioxidants are found in almost all plants, microorganisms, fungi and even in animal tissues (Yanishlieva-Maslarova 2001). In recent years, substances derived from the plant kingdom such as dried herbs and essential oils have been successfully used to reduce lipid oxidation in meat products (Estevez *et al.* 2005). Many plants having antioxidant properties are grown naturally in Turkey. Among them, myrtle, rosemary, nettle and lemon balm are the most important; they are cultivated and used for various purposes as traditional medicines. Many researchers have reported antioxidant effects of rosemary in meat products (Vareltzis *et al.* 1997; McCarthy *et al.* 2001a,b; Sanchez-Escalante *et al.* 2001; Nassu *et al.* 2003; Gimenez *et al.* 2004; Fernandez-Lopez *et al.* 2005). However, rosemary has been tested in meat systems, following two methods of application (dry powder and extracts in organic solvents). No studies have, however, appeared on the antioxidative effects of myrtle, nettle and lemon balm in meat products.

Consequently, our objective was to evaluate the effectiveness of myrtle, rosemary, nettle and lemon balm leaves on lipid oxidation and color stability of freezer-stored beef patties.

## MATERIALS AND METHODS

### Materials

Dried myrtle (*Myrtus communis myrtillus* L.), rosemary (*Rosmarinus officinalis* L.), nettle (*Urtica dioica*) and lemon balm (*Melissa officinalis* L.) leaves were purchased from a local market. Ground beef (moisture 64.07%, fat 15.09%) and backfat (moisture 10.60%, fat 87.39%) were purchased from a butcher shop. All chemicals used were of analytical grade.

### Preparation of the Plant Extracts

The dried leaves (myrtle, rosemary, nettle and lemon balm) were ground using a household coffee grinder. Plant powder (20 g) was mixed with 400 mL deionized water using a Waring blender (Waring Commercial Blender, Waring, Torrington, CT) for 15 min. The extract was then filtrated through filter paper (Whatman No. 1, Whatman International Ltd., Maidstone, UK) and used to prepare beef patties without further treatment.

## Preparation of Beef Patties

In the experiments, portions of uniform weight of the ground beef (about 1,500 g) were mixed with salt (1.5%). Five types of sample were prepared: (1) control sample (no extract added); (2) sample with addition of 10% (w/v) myrtle extract; (3) sample with addition of 10% rosemary extract; (4) sample with addition of 10% nettle extract; and (5) sample with addition of 10% lemon balm extract. Each group (1,500 g) was adjusted to a fat content of 20% by the addition of backfat, mixed thoroughly in a blender for 10 min, weighed into 25-g portions and shaped by hand. The beef patties were then wrapped in polyethylene cling film and stored for 120 days at  $-20 \pm 2^\circ\text{C}$ . Before analysis, the samples were defrosted in a refrigerator (Arcelik 475-T, Arcelik, Inc., Eskisehir, Turkey) at  $4^\circ\text{C}$  for 3 h. Oxidative and sensory changes were measured in triplicate samples on days 0, 30, 60, 90 and 120 of storage.

## pH, Peroxide Value (POV) and Thiobarbituric Acid-Reactive Substances (TBARS)

Samples were evaluated for pH using a digital pH meter (Cyberscan PC 510, Eutech Instruments, Singapore) after homogenizing a 10-g sample in 90 mL distilled water (Gokalp *et al.* 2001).

The content of hydroperoxides was estimated by determining the POV of the lipids expressed as meq  $\text{O}_2/\text{kg}$  lipid using the standard method of the Association of Official Analytical Chemists (AOAC 1990).

The thiobarbituric acid distillation method of Tarladgis *et al.* (1960) was used to determine the lipid oxidation. The rose-pink color produced by reaction between malondialdehyde (MDA) and 2-thiobarbituric acid ( $\text{C}_4\text{H}_4\text{N}_2\text{O}_2\text{S}$ ) was measured using a spectrophotometer (Helios Gamma, Thermo Spectronic, Madison, WI) at 538 nm. Lipid oxidation was expressed as 2-TBARS numbers (mg MDA/kg sample).

## Color

Beef patty surface color was measured using a Minolta CR 300 Chroma Meter (Minolta Camera Co., Osaka, Japan) which consisted of a measuring head (CR 300) with an 8-mm diameter measuring area and  $2^\circ$  standard observer with a C illuminant source, and a data processor (DP-301). The chroma meter was calibrated on the Hunterlab color space system using a white tile (Minolta calibration plate, No. 21733001,  $Y = 92.6$ ,  $x = 0.3136$ ,  $y = 0.3196$ ). Color was described as coordinates: lightness ( $L$ : 100 = white, 0 = black), redness ( $a \pm$  red-green) and yellowness ( $b \pm$  yellow-blue) for each patty. Three beef patties per each group were randomly selected and three readings were taken from each beef patty. Nine replicate measurements were

taken for each sample. The visual impression of color is formed from hue angle ( $H = \tan^{-1}[b/a]$ ) and chroma (saturation index) ( $c = [a^2 + b^2]^{1/2}$ ).

### **Sensory Evaluation**

Overall acceptance was evaluated monthly by a panel of 7–10 members from the Department of Food Engineering at Ondokuz Mayıs University in Turkey.

For sensory evaluation, each beef patty was cooked using an electric grill (Arcelik Mini Firin) at 300C (the distance between heat source and the samples was 4 cm) for 10 min (6 min one side and 4 min the other side). The cooked samples were cooled down at room temperature and served to panelists in random order. A 9-point hedonic scale, varying from dislike extremely (score 1) to like extremely (score 9), was used. Samples coded with random three-digit numbers were evaluated in each session and each judge was given approximately 20 g of each sample, served on plastic dishes. Water was allowed between samples. Assessments were carried out under fluorescence light. Five samples (each treatment from each replication) were evaluated in each session, presented separately. A total of three sessions were performed for each treatment. Samples were evaluated for overall acceptability with regard to color, taste and odor. Color was assessed on the raw patties while other sensory tests in cooked samples.

The overall acceptability was calculated taking into account color, taste and odor (each with 33.3%).

### **Statistical Analysis**

The data obtained from three replications were analyzed as a completely randomized design procedure using the general linear model procedure of the SPSS statistical package program (SPSS, Inc., Chicago, IL). The model included antioxidant extracts (control, myrtle, rosemary, nettle and lemon balm extract) and storage time (0, 30, 60, 90 and 120 days) as main effects, and all their interactions. The differences among means were tested for significance ( $P < 0.05$ ) by Duncan's multiple range test. The results of the statistical analyses were shown as mean values  $\pm$  SE in the tables for five storage times and five treatments.

## **RESULTS AND DISCUSSION**

### **pH, POV and TBARS**

The effect of the treatment and storage time on pH, POV and TBARS number of beef patties are shown in Table 1. As can be seen, antioxidant

TABLE 1.  
EFFECT OF TREATMENT AND STORAGE TIME ON pH, PEROXIDE VALUE (POV) AND  
THIOBARBITURIC ACID-REACTIVE SUBSTANCES (TBARS) VALUES OF BEEF PATTIES

Effect	pH	POV (meq O <sub>2</sub> /kg lipid)	TBARS (mg MDA/kg sample)
Treatment (A)	NS	**	**
Control	5.89 ± 0.06	5.19 ± 1.08 a	2.31 ± 0.32 a
Myrtle extract	5.87 ± 0.06	3.07 ± 0.40 d	0.96 ± 0.13 c
Rosemary extract	5.90 ± 0.08	3.47 ± 0.39 cd	0.97 ± 0.12 c
Nettle extract	5.94 ± 0.08	3.93 ± 0.55 bc	1.37 ± 0.19 b
Lemon balm extract	5.90 ± 0.06	4.01 ± 0.92 b	1.30 ± 0.17 b
Storage time (days) (B)	**	**	**
0	6.36 ± 0.04 a	1.31 ± 0.15 d	0.16 ± 0.01 d
30	5.76 ± 0.01 b	1.86 ± 0.14 c	1.31 ± 0.11 c
60	5.79 ± 0.02 b	3.15 ± 0.25 b	1.64 ± 0.20 b
90	5.82 ± 0.01 b	6.85 ± 0.59 a	1.78 ± 0.19 b
120	5.75 ± 0.04 b	6.51 ± 0.66 a	2.03 ± 0.22 a
A × B interaction	NS	**	**

Any two means in the same column having the same letters in the same section (treatment or storage time) are significantly different. Means based on five values.

\*\*  $P < 0.01$ .

NS, not significant ( $P > 0.05$ ).

MDA, malondialdehyde.

treatment had no significant ( $P > 0.05$ ) effect on pH, while pH decreased significantly ( $P < 0.01$ ) during storage period. The pH of beef patties was the highest at the beginning of storage ( $P < 0.05$ ). In the study made by Sanchez-Escalante *et al.* (2003), the antioxidant activity of lycopene-rich tomato-based products and peppers was investigated in beef patties. The pH levels of the samples were the highest at the beginning of storage, and the addition of any of the antioxidants, irrespective of concentration, did not significantly ( $P < 0.05$ ) affect the pH. According to Cheah and Hasim (2000), the addition of 1–10% galangal (*Alpinia galangal*) extract to minced beef was found not to affect the pH. Nassu *et al.* (2003) evaluated the oxidative stability of fermented goat meat sausages formulated with 0.025 and 0.050% levels of rosemary as a natural antioxidant. These two treatment levels had no significant effect on pH.

The primary products of lipid oxidation are hydroperoxides, which are turned to peroxides. Therefore, it seemed reasonable to determine the concentration of peroxide in the meat samples to study the extent of oxidation (Nawar 1996). As indicated in Table 1, both antioxidant treatment and storage time had statistically significant ( $P < 0.01$ ) effects on POV. The lowest POV observed in sample added with myrtle extract was 3.07 meq O<sub>2</sub>/kg lipid, but no significant difference with rosemary extract was observed ( $P > 0.05$ ). The highest POV was observed in control. The POVs of nettle- and lemon balm extract-added

samples were lower than that of the control ( $P < 0.05$ ). Myrtle and rosemary extracts showed the highest antioxidant effects and they achieved respectively 41 and 33% decrease in POV content with respect to control. The reduction of POV on samples containing nettle and lemon balm extracts was calculated as 24 and 23%, respectively. These results reveal that, hot-water extracts from myrtle, rosemary, nettle and lemon balm are effective in retarding the lipid oxidation of frozen stored beef patties. The antioxidant properties of these extracts are related to their phenolic contents. Phenolic antioxidants do not work as oxygen absorbers, rather they prevent formation of fatty free radicals, which react with or absorb oxygen in the autoxidation process, thus delaying the onset of the autoxidative process in fat or oil (Sherwin 1990). No reports have been found in the literature concerning the effects of the addition of plant extracts (myrtle, nettle and lemon balm) on meat oxidation. For this reason, the results of this study were compared with the findings of other researchers on rosemary and other plant extracts. Our results were in agreement with Junta-chote *et al.* (2006) who reported that dried galangal powder, dried holy basil powder, galangal extracts and holy basil extracts treated samples had significantly ( $P < 0.05$ ) lower peroxide values compared to control. Hassan and Fan (2005) also reported that the polyphenol extracts from cocoa leaves were able to stop peroxide formation in mechanically deboned chicken meats, and especially, the effect was more at higher concentrates (400 and 800 mg/kg).

In analyzed beef patties, POV increased steadily until the 90 days of storage ( $P < 0.05$ ) and then, began to decrease ( $P > 0.05$ ). This is probably because of the instability of hydroperoxides. It should also be mentioned that hydroperoxides begin to decompose as soon as they are formed. In the first stages of autoxidation, their rate of formation exceeds their rate of decomposition. The reverse takes place at the later stage (Nawar 1996). Similar to our results, increments in POV during storage of holy basil and galangal added cooked ground pork and polyphenol extracts from cocoa leaves added mechanically deboned chicken meat were reported by Hassan and Fan (2005) and Juntachote *et al.* (2006), respectively. The maximum acceptability limit of POV in fatty foods was reported as 25 meq  $O_2$ /kg lipid by Evranuz (1993). POV of beef patties did not reach this value in any of the storage months. An interaction ( $P < 0.01$ ) of the treatment and the storage time was observed for POV (Fig. 1). According to this figure, control and lemon balm extract added samples showed the highest increments while myrtle and rosemary added samples showed the lowest increments. The increases in peroxide value were higher between 30 and 60 days of storage for myrtle and rosemary extract added samples, and between 60 and 90 days of storage for control samples and nettle and lemon balm extract added samples. Except rosemary and lemon balm extracts, the POV of other treatments began to decline after 90 days. This is probably because of the instability of the peroxides.

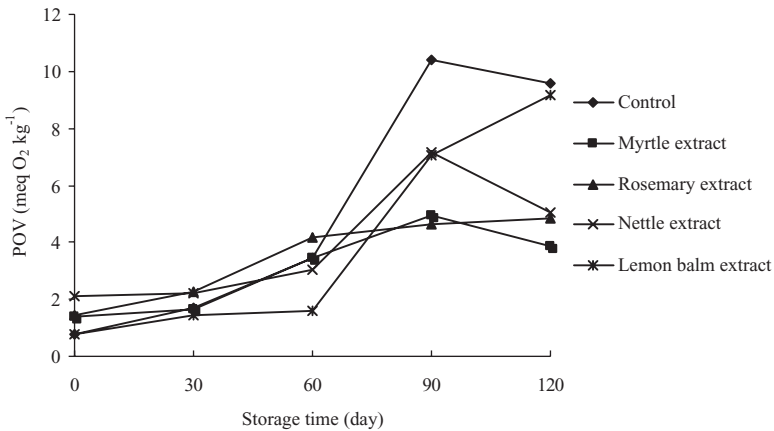


FIG. 1. EFFECT OF TREATMENT  $\times$  STORAGE TIME ON THE PEROXIDE VALUE (POV) OF BEEF PATTIES

TBARS test is one of the most widely used tests for evaluating the extent of lipid oxidation. In the progression of lipid peroxidation, the termination of the chain reactions results in the production of aldehydes, ketones and the other degradation components of oxidized fatty acids. The concentration of secondary products of lipid peroxidation, expressed as the quantity of the free MDA, is determined. These compounds, resulting from lipid oxidation, cause off-flavors in fatty foods (Nawar 1996). As shown in Table 1, treatment and storage time had statistically significant effect ( $P < 0.01$ ) on TBARS values of beef patties. Similar to POV, the lowest TBARS value occurred in the myrtle extract (0.96 mg MDA/kg sample) and rosemary extract (0.97 mg MDA/kg sample) added beef patties ( $P > 0.05$ ), while the highest TBARS value (2.31 mg MDA/kg sample) occurred in control ( $P < 0.05$ ). The antioxidant effectiveness of nettle and lemon balm extracts was lower than that of myrtle and rosemary extracts ( $P < 0.05$ ). According to these results, myrtle and rosemary extracts exhibited the highest antioxidant effects and showed 58% reduction in TBARS value with respect to control. This ratio was calculated as 44 and 41% for lemon balm and nettle extracts, respectively. All of the plant extracts used in this research showed antioxidant properties. These antioxidant activities have been attributed to the phenolic contents and chemical properties of these plants. The efficiency of various antioxidant extracts in inhibiting lipid oxidation throughout frozen storage was in the following order: myrtle extract = rosemary extract > lemon balm extract = nettle extract > control ( $P < 0.05$ ). Similar to these results, Fernandez-Lopez *et al.* (2005) reported that the TBARS values of cooked meat balls treated with rosemary oil extract, rosemary water extract, rosemary oil and water extract, garlic extract, lemon



extract and orange extract were significantly lower than those for control ( $P < 0.05$ ), and samples with rosemary extracts showed the lowest ( $P < 0.05$ ) TBARS values at each time of storage. Rey *et al.* (2005) also reported that the effect of cloudberry, willow herb and beetroot plant extracts on TBARS development of cooked pork patties was significant and the most potent antioxidant on stabilizing oxidation was cloudberry extract. The strong antioxidant effects of rosemary powder and extracts on meat products have been widely investigated (McCarthy *et al.* 2001b; Sanchez-Escalante *et al.* 2001). Moreover, the effects of various plant powders and extracts on TBARS value have been tested. Several researchers have shown the antioxidant potentials of plant extracts: fenugreek extracts delayed the induction period of oxidative rancidity of ground beef patties stored at 4C (Hettiarachchy *et al.* 1996); marjoram, wild marjoram and caraway showed the highest inhibitory effect on the TBARS value of minced chicken meat during frozen storage (El-Alim *et al.* 1999); tea catechins significantly ( $P < 0.01$ ) reduced lipid oxidation during the 10 days of refrigeration (4C) storage of cooked beef and chicken meat (Tang *et al.* 2001); tea catechins treatments greatly suppressed ( $P < 0.01$ ) lipid oxidation in raw meat patties (Mitsumoto *et al.* 2005); green tea and Pouchong tea were significantly more effective at inhibiting the formation of TBARS in bonito fillets than black tea (Lin and Lin 2005); polyphenol extract from cocoa leaves reduced TBARS formation of mechanically deboned chicken meat stored at 4C (Hassan and Fan 2005).

During frozen ( $-20^{\circ}\text{C}$ ) storage, lipid oxidation (TBARS) for beef patties increased steadily from 0.16 mg MDA/kg sample (day 1) to 2.03 mg MDA/kg sample (day 120) (Table 1). There were no significant changes in the TBARS between 60 and 90 days of storage ( $P > 0.05$ ). Various researchers observed increases in the amount of TBARS produced in meat and seafoods during the period of storage in the absence of any additives (Vareltzis *et al.* 1997; El-Alim *et al.* 1999; Cheah and Hasim 2000; McCarthy *et al.* 2001a,b; Sanchez-Escalante *et al.* 2003; Gimenez *et al.* 2004; Fernandez-Lopez *et al.* 2005; Hassan and Fan 2005; Lin and Lin 2005; Mitsumoto *et al.* 2005; Rey *et al.* 2005; Juntachote *et al.* 2006). Ferric hematin pigments are powerful catalysts of lipid oxidation in raw meat and model system emulsions. Ferric hematin pigments facilitate the transfer of electrons leading to increased rates of free radical formation. Sodium chloride accelerates oxidation of triglycerides, although the mechanism is not completely known. Also, grinding results in exposure of the labile lipid components to oxygen, and thus accelerates development of oxidative rancidity (Ladikos and Lougovois 1990).

An interaction ( $P < 0.01$ ) between treatment and storage time noted for TBARS values is shown in Fig. 2. According to the figure, there was an increase in TBARS values in both control samples and in those treated with antioxidants and this increase was the most rapid between the 0 and 30 days of

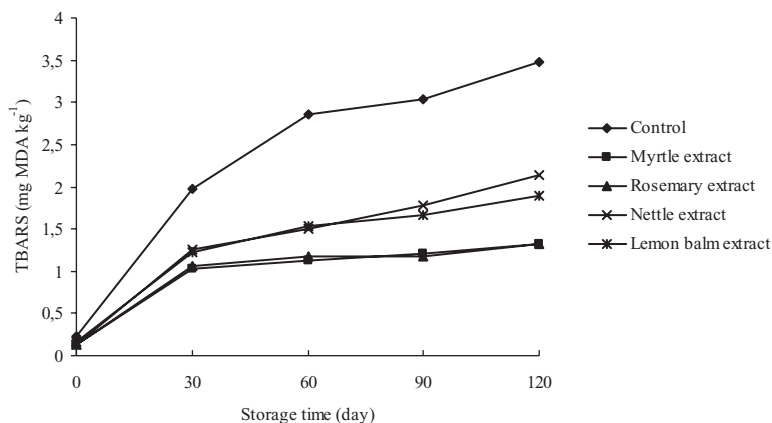


FIG. 2. EFFECT OF TREATMENT  $\times$  STORAGE TIME ON THE TBARS OF BEEF PATTIES

storage. Similarly, Varelziz *et al.* (1997) reported the highest increase in TBARS between 0 and 20 days of storage during the frozen storage ( $-18^{\circ}\text{C}$ ) of both filleted and minced frozen Mediterranean hake with or without antioxidants. Between the 30 and 120 days of storage, beef patties treated with myrtle and rosemary extracts had significantly slower TBARS increases than the other groups. The highest concentration of MDA was observed to occur in control samples as expected. According to these results, among the extracts studied, myrtle and rosemary extracts were significantly more effective in inhibiting the formation of TBARS.

## Color

Mean values for color components ( $L$ ,  $a$ ,  $b$ , hue angle and chroma) of beef patties are shown in Table 2. All except  $L$  values ( $P > 0.05$ ) were affected by the addition of plant extracts. Treatments showed significant effects on redness, hue angle and chroma ( $P < 0.01$ ), and yellowness ( $P < 0.05$ ). The highest value for parameter  $a$  (9.36) was obtained in myrtle extract added patties, while the lowest value (4.13) was observed in nettle extract added samples ( $P < 0.05$ ). No significant differences were found between control, rosemary and lemon balm extracts ( $P > 0.05$ ). The effects of plant extracts on chroma values of beef patties were similar to the effects on  $a$  value.  $b$  values of patties changed between narrow limits and the lowest value was recorded for myrtle extract added patties. The difference between myrtle and lemon balm extracts was statistically insignificant ( $P > 0.05$ ). Antioxidant added beef patties showed a trend opposite to  $a$  and chroma values. The highest value (67.27) was calculated for nettle extract, and the lowest value (44.57) for

TABLE 2.  
EFFECT OF TREATMENT AND STORAGE TIME ON COLOR PROPERTIES OF  
BEEF PATTIES

Effect	<i>L</i>	<i>a</i>	<i>b</i>	Hue angle	Chroma
Treatment (A)	NS	**	*	**	**
Control	42.04 ± 0.60	7.21 ± 0.43 b	10.05 ± 0.16 a	54.68 ± 1.85 b	12.46 ± 0.23 b
Myrtle extract	40.01 ± 0.44	9.36 ± 0.28 a	9.17 ± 0.14 b	44.57 ± 0.84 d	13.12 ± 0.26 a
Rosemary extract	41.39 ± 0.53	8.17 ± 0.31 b	9.72 ± 0.15 a	50.17 ± 1.07 c	12.73 ± 0.25 b
Nettle extract	40.87 ± 0.54	4.13 ± 0.31 c	9.74 ± 0.14 a	67.27 ± 1.57 a	10.63 ± 0.18 c
Lemon balm extract	40.83 ± 0.59	7.73 ± 0.36 b	9.62 ± 0.16 ab	51.46 ± 1.51 bc	12.40 ± 0.23 b
Storage time (days) (B)	NS	**	NS	**	**
0	41.57 ± 0.46	8.30 ± 0.58 a	9.74 ± 0.14	50.31 ± 2.33 c	12.94 ± 0.32 a
30	41.46 ± 0.76	7.61 ± 0.52 ab	9.91 ± 0.20	53.01 ± 2.16 bc	12.61 ± 0.34 a
60	40.37 ± 0.36	7.65 ± 0.54 ab	9.60 ± 0.15	52.12 ± 2.31 bc	12.41 ± 0.29 ab
90	40.78 ± 0.47	6.86 ± 0.52 bc	9.60 ± 0.16	55.05 ± 2.45 ab	11.94 ± 0.23 bc
120	40.95 ± 0.65	6.18 ± 0.57 c	9.45 ± 0.15	57.64 ± 2.58 a	11.44 ± 0.30 c
A × B interaction	NS	NS	NS	NS	NS

Any two means in the same column having the same letters in the same sections (treatment or storage time) are significantly different. Means based on five values.

\*  $P < 0.05$ .

\*\*  $P < 0.01$ .

NS, not significant ( $P > 0.05$ ).

myrtle extract. These results suggested that the addition of myrtle extract has protected color by retarding the formation of metmyoglobin in frozen stored beef patties. Myrtle also showed the same effect in lipid oxidation. The lowest *a* value of nettle extract was probably because of the color of this extract. Also, the oxidation of chlorophyll could cause this. Various researchers studied the effects of rosemary on color values of meats and meat products. Some of them were about the color protective effects of rosemary (Sanchez-Escalante *et al.* 2001; Estevez *et al.* 2005; Fernandez-Lopez *et al.* 2005) and the others (McCarthy *et al.* 2001a,b) were about the insignificant effects of rosemary on color properties as in this research. There were no research results in the literature about the effects of nettle and lemon balm extracts on color properties of meat and meat products.

*L* and *b* values showed no significant differences ( $P > 0.05$ ) among storage times, while redness, hue angle and chroma values were significantly different ( $P < 0.01$ ). The initial *a* value (8.30) decreased during storage, and reached to 6.18 at 120 days of storage. Chroma value showed a similar trend during storage. Except at 60 days of storage, hue angle increased. The decrement of *a* value during storage is probably caused by the oxidation of myoglobin. The decrease in *a* values of meat and meat products during storage has been previously reported (McCarthy *et al.* 2001a,b; Sanchez-Escalante *et al.* 2001, 2003; Estevez *et al.* 2005; Fernandez-Lopez *et al.* 2005). Estevez *et al.*

(2005) reported decreasing chroma values and increasing hue values during the storage of rosemary essential oil added frankfurters. Likewise, Sanchez-Escalante *et al.* (2003) determined increasing hue values of beef patties during storage.

### Sensory Evaluation

Table 3 shows the evaluation of sensory scores (color, taste, odor and overall acceptability) of beef patties added with extracts and storage time. Color, taste, odor and overall acceptability were affected significantly ( $P < 0.01$ ) with extract addition and with storage time. As for *a* value, samples with myrtle extract presented the highest color score, but beef patties with rosemary extract were not found to be different from patties subjected to myrtle extract treatment ( $P > 0.05$ ). Samples with nettle extract presented the lowest color score, but the differences between nettle extract and control and lemon balm extract were not significantly different ( $P > 0.05$ ). These results revealed that, myrtle and partly rosemary extracts prevented metmyoglobin formation, and therefore protected the color properties of beef patties. Taste and odor scores showed a similar trend and again, samples treated with myrtle and rosemary extracts were given the highest scores ( $P < 0.05$ ). These results show that addition of myrtle and rosemary extracts considerably delayed

TABLE 3.  
EFFECT OF TREATMENT AND STORAGE TIME ON SENSORY PROPERTIES OF  
BEEF PATTIES

Effect	Color	Taste	Odor	Overall acceptability
Treatment (A)	**	**	**	**
Control	6.44 ± 0.14 bc	6.33 ± 0.17 b	6.24 ± 0.15 b	6.34 ± 0.12 b
Myrtle extract	7.02 ± 0.16 a	6.73 ± 0.24 a	7.03 ± 0.20 a	6.93 ± 0.18 a
Rosemary extract	6.76 ± 0.16 ab	6.71 ± 0.21 a	6.82 ± 0.15 a	6.76 ± 0.15 a
Nettle extract	6.41 ± 0.14 c	6.25 ± 0.10 b	6.42 ± 0.13 b	6.36 ± 0.08 b
Lemon balm extract	6.51 ± 0.15 bc	6.22 ± 0.18 b	6.33 ± 0.18 b	6.35 ± 0.13 b
Storage time (days) (B)	**	**	**	**
0	7.34 ± 0.14 a	7.37 ± 0.19 a	7.15 ± 0.22 a	7.29 ± 0.16 a
30	6.72 ± 0.12 b	6.53 ± 0.16 b	6.53 ± 0.21 b	6.59 ± 0.13 b
60	6.55 ± 0.10 b	6.31 ± 0.10 b	6.45 ± 0.13 b	6.44 ± 0.07 bc
90	6.43 ± 0.10 b	6.19 ± 0.08 b	6.44 ± 0.10 b	6.35 ± 0.06 c
120	6.11 ± 0.13 c	5.83 ± 0.13 c	6.27 ± 0.12 b	6.07 ± 0.08 d
A × B interaction	NS	NS	NS	NS

Any two means in the same column having the same letters in the same sections (treatment or storage time) are significantly different. Means based on five values.

\*\*  $P < 0.01$ .

NS, not significant ( $P > 0.05$ ).

rancidity, because POV and TBARS values also supported this conclusion. Overall acceptability was calculated considering color, taste and odor scores and samples with myrtle and rosemary extracts presented highest scores, respectively 6.93 and 6.76 ( $P < 0.05$ ). No significant differences ( $P > 0.05$ ) with respect to the rest of the treatments were found. These results agreed with those obtained by Sanchez-Escalante *et al.* (2001) who found that rosemary extract was the most effective in inhibiting oxidation of both lipid and myoglobin, and contributed to maintaining desirable sensory characteristics of fresh patties and therefore extending their shelf life. Similarly, Fernandez-Lopez *et al.* (2005) reported that sensory analysis results, particularly aroma and acceptability scores, indicated significant advantages in using rosemary extract in rancidity-susceptible meat products.

As shown in Table 3, color, taste, odor and overall acceptability scores of beef patties numerically decreased during storage and the same trend was observed in color and taste scores. The variations in the sensory properties of antioxidant-added meat products were examined by Gimenez *et al.* (2004) and Fernandez-Lopez *et al.* (2005) during storage. Common results were the decrease of sensory properties dependent on storage period. Therefore, the results of this research were in good agreement with those of other researchers.

## CONCLUSION

The addition of hot-water extracts of myrtle leaves at 10% ratio to beef patties stored at  $-20 \pm 2^\circ\text{C}$  delayed lipid oxidation and provided color protection. In addition, the application of rosemary extracts could be useful to control lipid oxidation. These observations deserve to be explored in more detail in the future.

## ACKNOWLEDGMENT

This work was financially supported by Ondokuz Mayıs University Research Foundation (MF.082).

## REFERENCES

- AOAC. 1990. *Official Methods of Analysis*, 15th Ed., Association of Official Analytical Chemists, Arlington, VA.
- BALEV, D., VULKOVA, T., DRAGOEV, S., ZLATANOV, M. and BAHTCHEVANSKA, S. 2005. A comparative study on the effect of

- some antioxidants on the lipid and pigment oxidation in dry-fermented sausages. *Int. J. Food Sci. Technol.* 40, 977–983.
- BEKHIT, A.E.D., GEESING, G.H., ILIAN, M.A., MORTON, J.D. and BICKERSTAFFE, R. 2003. The effects of natural antioxidants on oxidative processes and methmyoglobin reducing activity in beef patties. *Food Chem.* 81, 175–187.
- CHEAH, P.B. and HASIM, N.H.A. 2000. Natural antioxidant extract from galangal (*Alpinia galanga*) for minced beef. *J. Sci. Food Agric.* 80, 1565–1571.
- EL-ALIM, S.S.L.A., LUGASI, A., HOVARI, J. and DWORSCHAK, E. 1999. Culinary herbs inhibit lipid oxidation in raw and cooked minced meat patties during storage. *J. Sci. Food Agric.* 79, 277–285.
- ESTEVEZ, M., VENTANAS, S. and CAVA, R. 2005. Protein oxidation in frankfurters with increasing levels of added rosemary essential oil: Effect of color and texture deterioration. *J. Food Sci.* 70, 427–432.
- EVRAUZ, O. 1993. The effects of temperature and moisture content on lipid peroxidation during storage of unblanched salted roasted peanuts: Shelf life studies of unblanched salted roasted peanuts. *Int. J. Food Sci. Technol.* 28, 193–199.
- FERNANDEZ-LOPEZ, J., ZHI, N., ALESON-CARBONELL, L., PEREZ-ALVAREZ, J.A. and KURI, V. 2005. Antioxidant and antibacterial activities of natural extracts: Application in beef meatballs. *Meat Sci.* 69, 371–380.
- FOEGEDING, E.A., LANIER, T.C. and HULTIN, H.O. 1996. Characteristics of edible muscle tissues. In *Food Chemistry*, 4th Ed. (O.R. Fennema, ed.) pp. 725–790, Marcel Dekker, Inc., New York, NY.
- GIMENEZ, B., RONCALES, P. and BELTRAN, J.A. 2004. The effects of natural antioxidants and lighting conditions on the quality characteristics of gilt-head sea bream fillets (*Sparus aurata*) packaged in a modified atmosphere. *J. Sci. Food Agric.* 84, 1053–1060.
- GOKALP, H.Y., OCKERMAN, H.W., PLIMPTON, R.F. and HARPER, W.J. 1983. Fatty acid of neutral and phospholipids, rancidity scores and TBA values as influenced by packaging and storage. *J. Food Sci.* 48, 829–834.
- GOKALP, H.Y., KAYA, M., TULEK, Y. and ZORBA, O. 2001. *Et Urunlerinde Kalite Kontrolu ve Laboratuvar Uygulama Klavuzu*, Yayın No: 751, Ataturk University, Erzurum, Turkey.
- HASSAN, O. and FAN, L.S. 2005. The anti-oxidation potential of polyphenol extract from cocoa leaves on mechanically deboned chicken meat. *LWT-Food Sci. Technol.* 38, 315–321.
- HETTIARACHCHY, N.S., GLENN, K.C., GNANASAMBANDAM, R. and JOHNSON, M.G. 1996. Natural antioxidant extract from fenugreek

- (*Trigonella foenumgraecum*) for ground beef patties. *J. Food Sci.* 61, 516–519.
- JUNTACHOTE, T., BERGHOFER, E., SIEBENHANDL, S. and BAUER, F. 2006. The oxidative properties of Holy basil and Galangal in cooked ground pork. *Meat Sci.* 72, 446–456.
- LADIKOS, D. and LOUGOVOIS, V. 1990. Lipid oxidation in muscle foods: A review. *Food Chem.* 35, 295–314.
- LIN, C.C. and LIN, C.S. 2005. Enhancement of the storage quality of frozen bonito fillets by glazing with tea extracts. *Food Control* 16, 169–175.
- MCCARTHY, T.L., KERRY, J.P., KERRY, J.F., LYNCH, P.B. and BUCKLEY, D.J. 2001a. Evaluation of the antioxidant potential of natural food/plant extracts as compared with synthetic antioxidants and vitamin E in raw and cooked pork patties. *Meat Sci.* 57, 45–52.
- MCCARTHY, T.L., KERRY, J.P., KERRY, J.F., LYNCH, P.B. and BUCKLEY, D.J. 2001b. Assessment of the antioxidant potential of natural food and plant extracts in fresh and previously frozen pork patties. *Meat Sci.* 57, 177–184.
- MITSUMOTO, M., O'GRADY, M.N., KERRY, J.P. and BUCKLEY, D.J. 2005. Addition of tea catechins and vitamin C on sensory evaluation, colour and lipid stability during chilled storage in cooked or raw beef and chicken patties. *Meat Sci.* 69, 773–779.
- NASSU, R.T., GONCALVES, L.A.G., SILVA, M.A.A.P. and BESERRA, F.J. 2003. Oxidative stability of fermented goat meat sausage with different levels of natural antioxidant. *Meat Sci.* 63, 43–49.
- NAWAR, W.W. 1996. Lipids. In *Food Chemistry*, 4th Ed. (O.R. Fennema, ed.) pp. 139–245, Marcel Dekker, Inc., New York, NY.
- POKORNY, J. 2001. Introduction. In *Antioxidants in Food, Practical Applications*, 1st Ed. (J. Pokorny, N. Yanishlieva and M. Gordon, eds.) pp. 1–3, Woodhead Publishing Ltd, Cambridge, U.K.
- REY, A.I., HOPIA, A., KIVIKARI, R. and KAHKONEN, M. 2005. Use of natural food/plant extracts: Cloudberry (*Rubus Chamaemorus*), beetroot (*Beta Vulgaris* “*Vulgaris*”) or willow herb (*Epilobium angustifolium*) to reduce lipid oxidation of cooked pork patties. *LWT-Food Sci. Technol.* 38, 363–370.
- SANCHEZ-ESCALANTE, A., DJENANE, D., TORRESCANO, G., BELTRAN, J.A. and RONCALES, P. 2001. The effects of ascorbic acid, taurine, carnosine and rosemary powder on colour and lipid stability of beef patties packaged in modified atmosphere. *Meat Sci.* 58, 421–429.
- SANCHEZ-ESCALANTE, A., TORRESCANO, G., DJENANE, D., BELTRAN, J.A. and RONCALES, P. 2003. Stabilisation of colour and odour of beef patties by using lycopene-rich tomato and peppers as a source of antioxidants. *J. Sci. Food Agric.* 83, 187–194.

- SHERWIN, E.R. 1990. Antioxidants. In *Food Additives*, 1st Ed. (A.L. Branen, P.M. Davidson and S. Salminen, eds.) pp. 139–191, Marcel Dekker, Inc., New York, NY.
- TANG, S., KERRY, J.P., SHEEHAN, D. and BUCKLEY, D.J. 2001. A comparative study of the catechins and  $\alpha$ -tocopherol as antioxidants in cooked beef and chicken meat. *Eur. Food Res.* 213, 286–289.
- TARLADGIS, B.G., WATTS, B.M. and YOUNATHAN, M.T. 1960. A distillation method for the quantitative determination of malonaldehyde in rancid foods. *J. Am. Oil Chem. Soc.* 37, 44–48.
- VARELTZIS, K., KOUFIDIS, D., GAVRIILIDOU, E., PAPAVERGOU, E. and VASILIADOU, S. 1997. Effectiveness of natural rosemary (*Rosmarinus officinalis*) extract on the stability of filleted and minced fish during frozen storage. *Z. Lebensm.-Unters.-Forsch. A* 205, 93–96.
- YANISHLIEVA-MASLAROVA, N.V. 2001. Inhibiting oxidation. In *Antioxidants in Food, Practical Applications*, 1st Ed. (J. Pokorny, N. Yanishlieva and M. Gordon, eds.) pp. 20–68, Woodhead Publishing Ltd, Cambridge, U.K.