Efficacy of olive leaf extract for enhancing quality of beef cubes

K.K. Aytul[1], F. Korel[2*], D.K. Arserim-Uçar[2], İ. Uysal[2] & O. Bayraktar[3]
[1]Biotechnology and Bioengineering Program, İzmir Institute of Technology, Urla, İzmir, Turkey.
[2]Food Engineering Department, İzmir Institute of Technology, Urla, İzmir, Turkey.
[3]Chemical Engineering Department, İzmir Institute of Technology, Urla, İzmir, Turkey.
[*]E-mail: figenkorel@iyte.edu.tr

Abstract

In this study antioxidant and antimicrobial effects of olive leaf extract on the quality of beef cubes were investigated. The beef cubes were immersed into solutions containing 0, 1, 2, and 3% olive leaf extract (w/v) in 1:1 ratio (beef:water, w/v) for 20 hrs. Samples were then placed in Ziploc® plastic storage bags and stored at 4 ºC for 9 days. Meat spoilage (total viable, coliform, and lactic acid bacteria counts), moisture, pH, lightness, chroma, hue angle, and oxidative stability were determined. The results indicated the good potential of using olive leaf extract to enhance microbial and oxidative quality of cold stored beef cubes.
Keywords: meat quality, olive leaf extract, oleuropein

Introduction

Deterioration of meat products is mainly caused by lipid oxidation and microbial spoilage. The use of antioxidants and antimicrobials provides an effective way for preservation of meat products against lipid oxidation and microbial spoilage. An increased interest has been directed towards plant-based extracts as a source of phenolic antioxidants (Skerget et al. 2005) and antimicrobials. Generally, rosemary, oregano, and pimento are considered to be among the most potent antioxidants and antimicrobials (Estévez et al. 2006; Solomakos et al. 2007). Olive fruits, oil, and leaves also have antioxidant and antimicrobial properties due to their phenolic compounds, particularly oleuropein (McDonald et al. 2001).

The objective of this study was to determine the antioxidant and antimicrobial effects of olive leaf extract (OLE) on the quality of beef cubes stored at 4ºC up to 9 days.

Materials and methods

Beef (frozen and vacuum-packed) was obtained from Pınar Meat Co. (İzmir, Turkey) and they were cut into approximately 1.5x1.5 cm cubes immediately after thawing. Then they were divided into 4 groups (control and treated samples). Control samples were immersed into distilled water and the other 3 groups of samples were immersed into solutions containing 1%, 2% or 3% olive leaf extract (OLE) (w/v) in 1:1 ratio (beef:distilled water w/v) for 20 hrs at 4ºC. Samples were then placed in Ziploc® bags and stored at 4ºC for 9 days. The experiment was done in triplicate.

The oleuropein content of OLE was measured using HPLC (Hewlett-Packard series HP1100 equipped with a diode array dedector) (Garcia et al. 2000).

Moisture content of control and treated samples was analyzed in triplicate by AOAC method at days 1, 3, 6, and 9 (AOAC 1999). The pH of samples was measured in duplicate by a pH meter (Hanna Instruments, Portugal). Thiobarbituric acid, TBA as mg malonaldehyde equiv./kg (Tarladgis et al. 1960) was measured in control and treated samples in triplicate at days 1, 3, 6, and 9.

Surface color attributes of control and treated beef cubes were determined by color machine vision system (ECS, Inc., USA) at days 0, 1, 3, 6, and 9. Three samples from each treatment were placed in a light box and a 24-bit image was taken with a charged couple device (CCD) video camera, located inside the light box. The average L*, a*, b* values of all the pixels representing the sample were calculated. Chroma [$C^* = (a^{*2}+b^{*2})^{0.5}$] and hue angle [$h^* = \arctan (b^*/a^*)$] were also calculated (Little 1975).

Samples (10 g) were aseptically mixed with 90 ml of 0.1% peptone water and homogenized using a stomacher (Bagmixer® 400, Interscience, France). Mixtures were serially diluted (1:10) in 0.1% peptone water. Sample dilutions were plated and incubated to determine the microbial counts of total viable bacteria on plate count agar (PCA) at 30ºC/48 hrs and coliforms on violet red bile agar at 37ºC/24 hrs (VRBA). Lactic acid bacteria were enumerated using DeMan, Rogosa and Sharp agar (MRS) and Petri plates were incubated at 37ºC/48 hrs in a CO_2 incubator (5% CO_2 and 50% humidity). Microbial counts were expressed as $\log_{10}$ cfu/g of sample.

Data were analyzed using Statistical Analysis System (SAS Institute, Inc., Cary, NC, USA). General Linear Model (GLM) was used to evaluate treatment and storage as fixed effects. Duncan's multiple range

test was used to determine significant differences among treatments at each storage time and also among storage times at each treatment at $p<0.05$.

Results and discussion

The oleuropein content of OLE was 9.4 mg/ml. No significant differences in moisture content of the control and treated samples differing in OLE concentration were found among the treatments; however, storage time had a significant effect within the same treatment ($p<0.05$). The pH of the samples varied from 6.11±0.02 to 6.84±0.02. Changes in TBA values of the control and treated samples during storage are given in Table 1. The TBA values of control and treated samples were 0.63±0.19-1.00±0.02 mg malonaldehyde equiv./kg at day 1. The TBA values increased during storage due to the lipid oxidation. The samples treated with different concentrations of OLE had lower TBA values than of control samples at day 9. Treatment and storage time had significant effects on TBA values ($p<0.05$).

Table 1. TBA values as mg malonaldehyde equiv./kg for beef cubes treated with different concentrations of olive leaf extract during 9 days of storage at 4ºC

Treatment	Storage Time (days)			
	1	3	6	9
Control	$0.63\pm0.19^{c,x}$	$1.34\pm0.23^{b,x}$	$1.47\pm0.30^{b,x}$	$4.47\pm0.13^{a,w}$
%1 OLE	$0.70\pm0.27^{c,wx}$	$1.81\pm0.16^{b,w}$	$1.98\pm0.17^{b,w}$	$3.12\pm0.10^{a,y}$
%2 OLE	$0.77\pm0.09^{b,wx}$	$0.84\pm0.01^{b,y}$	$0.94\pm0.18^{b,y}$	$2.32\pm0.08^{a,z}$
%3 OLE	$1.00\pm0.02^{d,w}$	$1.77\pm0.03^{b,w}$	$1.27\pm0.02^{c,xy}$	$3.58\pm0.16^{a,x}$

$^{a-d}$:Means having different letters within each treatment denote significant difference at $p<0.05$.
$^{w-z}$:Means having different letters within each storage time denote significant difference at $p<0.05$.
Data are mean values ± S.D. (n=3)

The average L*, a*, b*, chroma and hue angle values for all samples during 9 days of storage are given in Table 2. The treatment and storage time had significant effects on all color attributes ($p<0.05$). Control samples had higher L*, a*, b*, and chroma values than of treated samples and as the concentration of OLE increased, the L*, a*, b*, and chroma values of the samples (stored up to 6 days) decreased.

Table 2. Color analysis for beef cubes treated with different concentrations of olive leaf extract during 9 days of storage at 4ºC

Color values	Treatments	Storage Time (days)				
		0	1	3	6	9
L* (lightness)	Control	51.59 ± 0.46^{e}	$62.80\pm0.39^{b,w}$	$66.46\pm0.01^{a,w}$	$61.46\pm0.16^{c,w}$	$56.83\pm0.33^{d,wy}$
	1% OLE	51.59 ± 0.46^{c}	$61.62\pm0.16^{ab,w}$	$63.45\pm2.51^{a,wy}$	$58.10\pm0.15^{b,y}$	$58.50\pm0.13^{b,w}$
	2% OLE	51.59 ± 0.46^{c}	$61.61\pm0.91^{a,w}$	$61.74\pm0.04^{a,yx}$	$56.04\pm0.07^{b,x}$	$55.09\pm0.52^{b,yx}$
	3% OLE	51.59 ± 0.46^{d}	$56.39\pm0.38^{b,y}$	$59.00\pm0.49^{a,x}$	$54.34\pm0.08^{c,z}$	$53.08\pm1.32^{c,x}$
a* (redness)	Control	41.90 ± 1.76^{c}	$44.42\pm0.23^{ab,w}$	$44.36\pm0.06^{ab,w}$	$46.32\pm0.09^{a,w}$	$42.36\pm0.08^{bc,w}$
	1% OLE	41.90 ± 1.76^{c}	$42.19\pm0.12^{b,x}$	$45.09\pm1.48^{a,w}$	$38.97\pm0.53^{d,y}$	$37.55\pm0.37^{d,y}$
	2% OLE	41.90 ± 1.76^{c}	$42.89\pm0.18^{b,y}$	$44.00\pm0.05^{a,w}$	$31.32\pm0.23^{e,x}$	$35.26\pm0.16^{d,x}$
	3% OLE	41.90 ± 1.76^{a}	$35.96\pm0.13^{b,z}$	$37.17\pm0.54^{b,y}$	$27.93\pm0.03^{c,z}$	$36.60\pm0.85^{b,y}$
b* (yellowness)	Control	25.05 ± 2.02^{c}	$32.58\pm0.45^{b,y}$	$44.30\pm0.02^{a,w}$	$33.05\pm0.40^{b,w}$	$23.07\pm0.30^{c,y}$
	1% OLE	25.05 ± 2.02^{d}	$32.63\pm0.01^{b,y}$	$39.55\pm1.34^{a,y}$	$29.56\pm0.51^{c,y}$	$20.52\pm0.34^{e,x}$
	2% OLE	25.05 ± 2.02^{d}	$35.87\pm0.91^{b,w}$	$40.57\pm0.12^{a,y}$	$28.93\pm0.01^{c,y}$	$21.91\pm0.07^{e,y}$
	3% OLE	25.05 ± 2.02^{d}	$29.67\pm0.20^{b,x}$	$33.73\pm0.60^{a,x}$	$27.57\pm0.08^{c,x}$	$24.57\pm0.84^{d,w}$
Chroma (intensity)	Control	48.82 ± 2.55^{c}	$55.09\pm0.09^{b,w}$	$62.69\pm0.06^{a,w}$	$56.90\pm0.16^{b,w}$	$48.24\pm0.07^{c,w}$
	1% OLE	48.82 ± 2.55^{c}	$53.33\pm0.10^{b,y}$	$59.99\pm0.22^{a,y}$	$48.91\pm0.73^{c,y}$	$42.79\pm0.16^{d,yx}$
	2% OLE	48.82 ± 2.55^{c}	$55.91\pm0.73^{b,w}$	$59.84\pm0.05^{a,y}$	$42.64\pm0.18^{d,x}$	$41.51\pm0.18^{e,x}$
	3% OLE	48.82 ± 2.55^{d}	$46.62\pm0.22^{b,x}$	$50.19\pm0.00^{a,x}$	$39.25\pm0.04^{d,z}$	$44.08\pm1.17^{c,y}$
Hue angle	Control	30.77 ± 0.86^{c}	$36.13\pm0.53^{b,x}$	$45.00\pm0.00^{a,w}$	$35.56\pm0.27^{b,z}$	$28.59\pm0.31^{d,x}$
	1% OLE	30.77 ± 0.86^{c}	$37.60\pm0.00^{b,y}$	$41.32\pm1.82^{a,y}$	$37.23\pm0.00^{b,x}$	$28.59\pm0.31^{d,x}$
	2% OLE	30.77 ± 0.86^{c}	$39.86\pm0.71^{b,w}$	$42.61\pm0.00^{a,wy}$	$42.77\pm0.22^{a,y}$	$31.80\pm0.00^{c,y}$
	3% OLE	30.77 ± 0.86^{e}	$39.52\pm0.24^{c,w}$	$42.30\pm0.88^{b,wy}$	$44.57\pm0.21^{a,w}$	$34.02\pm0.28^{d,w}$

$^{a-e}$:Means having different letters within each treatment denote significant difference at $p<0.05$.
$^{w-z}$:Means having different letters within each storage time denote significant difference at $p<0.05$.
Data are mean values ± S.D. (n=3)

Total viable bacteria and coliform counts for the samples treated with 2 and 3% OLE were lower than of control and 1% OLE treated samples (Figure 1). The microbial growth decreased for the samples treated with OLE compared to control samples. However, the total viable counts for all samples exceeded the spoilage limit which was 10^6-10^7 cfu/g. Different concentrations of OLE did not show any significant effect on lactic acid bacteria counts.

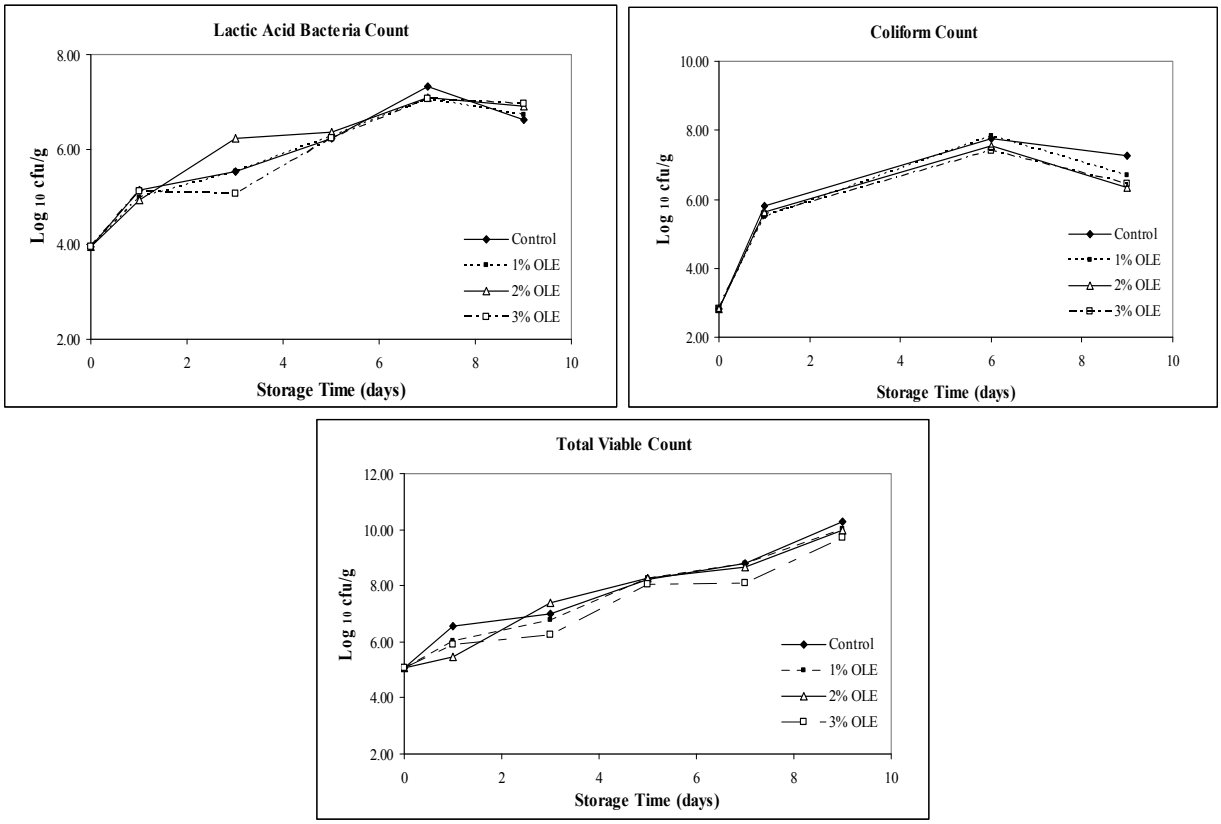

Figure 1. Total viable, coliform and lactic acid bacteria counts of beef cubes treated with different concentrations of olive leaf extract (OLE) during 9 days of storage at 4ºC.

The results clearly indicated that using of 2 and 3 % OLE had the beneficial effect in controlling the microbial load of beef cubes during 9 days of storage at 4ºC. Two percent OLE treatment applied to beef cubes also delayed the oxidative deterioration compared to the other samples. On the other hand, no beneficial effects of OLE treatment were determined on surface color attributes, moisture content and pH values of beef cubes.

References

AOAC, 1999. Official Methods of Analysis of AOAC International. 16th ed. 5th revision. Gaithersburg, MD.

Estévez, M., Cava, R., 2006. Effectiveness of rosemary essential oil as an inhibitor of lipid and protein oxidation: Contradictory effects in different types of frankfurters. Meat Science 72, 348-355.

Garcia, O.B., Castillo, J., Lorente, J., Ortuno, A., Del Rio, J.A. 2000. Antioxidant activity of phenolics extracted from *Olea europaea* L. leaves. Food Chemistry 68, 457-462.

Little, A.C. 1975. Off on a tangent. Journal of Food Science 40, 410.

Tarladgis, B.G., Watts, B.M., Younathan, M.S., Dugan, L.Jr. 1960. A distillation method for the quantitative determination of malonaldehyde in rancid foods. Journal of American Oil Chemists Society, 37, 44-48.

McDonald, S., Prenzler, P.D., Antholovich, M., and Robards, K. 2001. Phenolic content and antioxidant activity of olive extracts. Food Chemistry 73, 73-84.

Skerget, M. Kotnik P., Hadolin, M., Hras A.R., Knez Z., 2005. Phenols, proanthocyanidins, flavons and flavonols in some plant materials and their antioxidant activities. Food Chemistry 89, 191-198.

Solomakos, N., Govaris, A., Koidis, P., Botsoglou, N., 2007. The antimicrobial effect of thyme essential oil, nisin and their combination against *Escherichia coli* O157:H7 in minced beef during refrigerated storage. Meat Science (in press).