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Investigation of the effect of lavender (*Lavandula angustifolia* Mill.) essential oil on microbiological, physicochemical, and sensorial properties of meatballs during shelf-life, and its inhibitory effect on *Escherichia coli* O157:H7

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Abstract

The present work aimed at investigating the effect of lavender (*Lavandula angustifolia* Mill.) essential oil on the microbiological, physicochemical, and sensorial properties of meatballs, and elucidating its antimicrobial effect on the meatballs contaminated with *Escherichia coli* O157:H7. The essential oil as determined via gas chromatography mass spectrometry (GC-MS) contained two major components, linalool and linalyl acetate, at 37.023 and 28.651%, respectively. The antioxidant activity test which was performed via 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity test, revealed that the essential oil had moderate capacity. The antibacterial activity of the essential oil was investigated against *E. coli* O157:H7, and the minimum inhibition concentration (MIC) was determined to be 6.4 µL/mL. Six different test groups were formed based on the concentrations of *E. coli* O157:H7 (0 and 10^8 CFU/mL) and essential oil (0, 6.4, and 12.8 µL/mL). Although essential oil had inhibitory effect against TAMB, coliforms, yeasts and moulds, *Staphylococcus aureus*, *E. coli*, and especially *E. coli* O157:H7 in the microbiological analyses, no effect was observed on the physicochemical properties of the meatballs. Approximately, 3 log decrease was observed in the *E. coli* O157:H7 levels when essential oil was added at a concentration of two-fold MIC value. It was observed that the addition of essential oil to meatballs did not increase the lipid oxidation level as much as the control group. Meatballs that contained essential oil at MIC value achieved the highest general acceptability scores at the end of their storage periods.

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Introduction

Meatballs as processed products are more vulnerable to contamination since during mincing, a critical process in the preparation of meatballs, it is prone to cross contamination due to increased surface area and human intervention. The risk of contamination makes this food a severe concern for public health (Fernandez-Lopez *et al*., 2005). *Escherichia coli* O157:H7 in the Shiga toxin-producing *Escherichia coli* (STEC) group is one of the most important foodborne pathogens mainly related to meat and meat products (Pennington, 2010).

The use of synthetic preservatives is an effective way preferred by the food industry in controlling foodborne infections. However, new products that can ensure food safety are required due to consumers' raised awareness regarding health effects, and increased antibiotic resistance (Man *et al*., 2019). Studies have proved that essential oils have strong antimicrobial effects, and can be used to replace synthetic products in food preservation (Gavanji *et al*., 2015).

Lavender essential oil derived from *Lavandula angustifolia* Mill., which is a genus in the family Lamiaceae, has long been used in cosmetics and medicines for its antimicrobial, antidepressant, and sedative effects (Cavanagh and Wilkinson, 2002) as it contains aldehydes, ketones, phenols, and terpenes (*i.e.*, linalool, linalyl acetate, and camphor) (Ceylan and Fung, 2004).

Lavender essential oil has also been used as preservative in a variety of foods (*i.e.*, vegetables,

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rice, fruits, dairy, meat, and fish products) (Burt, 2004) to prolong their shelf-life (Lucera *et al*., 2012). However, its dominant aroma may limit its use in food (Marín *et al*., 2016).

Therefore, the aim of the present work was to investigate the effects of *L. angustifolia* essential oil on the microbiological, physicochemical, and sensorial parameters of meatballs to obtain a safe, and novel meat-based functional food which would not put public health at risk.

Materials and methods

Preparation of E. coli O157:H7 strain inoculum

E. coli O157:H7 strain (ATCC43895), which was artificially inoculated into the homemade meatball samples, was obtained from the bacterial culture collection of Department of Food Hygiene and Technology, Mehmet Akif Ersoy University, Türkiye. Bacterial strain was cultured on Mueller-Hinton agar (MHA, OR-BAK, OR-PET 35) at 37°C overnight. A representative colony was inoculated into Mueller-Hinton broth (MHB, Merck 70192) and incubated at 37°C for 24 h. Working fresh inoculum (about 10^8 CFU/mL) was obtained based on 0.5 McFarland standard (Ben Hsouna and Hamdi, 2012).

Plant material

Essential oil was obtained by water steam distillation method from *L. angustifolia* which was cultivated in Burdur (37°43'07.3"N, 30°04'57.8"E) province and its surroundings. To prepare the stock solution, essential oil was dissolved in 10% dimethyl sulfoxide (DMSO) (Sigma Aldrich®-Química, S.A.) to a final concentration of 51.2 µL/mL, and stored for further use.

Characterisation of Lavandula angustifolia essential oil

The composition of *L. angustifolia* essential oil (Table 1) was determined via GC-MS analysis performed via AGILENT7 GC 890A (Agilent Technologies), equipped with a capillary PWAX 52 CB column (50 m length, 0.25 mm diameter, 0.2 mm film thickness) and coupled with a mass detector (AGILENT MSD 5975C; Agilent Technologies). The operating conditions were as follows: injector and detector temperatures, 240 and 250°C. The oven was heated at a gradient mode starting with an initial temperature of 60°C and continued for 2 min, followed by an increase to 220°C at a rate of 2°C/min, and the conditions were kept constant at this temperature for 20 min (Baydar *et al*., 2013).

Determination of the antioxidant activity of Lavandula angustifolia essential oil

Radical scavenging activity was determined following the method described by Brand-Williams *et al*. (1995) with slight modifications. Briefly, 0.1 mM DPPH (2,2-diphenyl-1-picrylhydrazyl) solution was prepared in ethanol. Next, 3 mL of the prepared DPPH solution was added onto different volumes of the essential oil (0.1 - 0.5 mL), and the final volumes of which were made 1 mL with ethanol. Then, the solutions were incubated at room temperature for 30 min in the dark. Ethanol was used as blank solution, and ascorbic acid as standard. Absorbance was measured at 517 nm via a UV-visible spectrophotometer (PerkinElmer, L60000CC, Massachusetts, UK), and DPPH radical scavenging activity was determined as percent inhibition (I %) using Eq. 1:

$$I\ (\%) = [(A_0 - A_1) / A_0] \times 100 \qquad \text{(Eq. 1)}$$

where, A_0 = absorbance of the control group, and A_1 = absorbance of the sample.

The IC_{50} value, expressed as µg/mL, was determined using the value found via linear interpolation of the inhibition values. The tests were performed in triplicate.

Antimicrobial activity of Lavandula angustifolia essential oil via disk diffusion method

Mueller-Hinton agar (MHA, OR-BAK, OR-PET 35) plates were inoculated with 100 µL (10^8 CFU/mL) of the bacterial suspension. Then, sterile filter paper discs in 6 mm diameter were impregnated with 5 µL of *L. angustifolia* essential oil dissolved in DMSO using a micropipette, and placed onto the inoculated MHA plates. After incubation at 37°C for 24 h, inhibition zone diameters were determined following a method previously described (Djenane *et al*., 2011). For negative control, DMSO and essential oil without bacterial inoculum was used. For positive control, disks containing gentamicin (OXOİD, 10 µg, 10538623) was used.

Minimum inhibitory concentration (MIC)

The antimicrobial activity of *L. angustifolia*

Table 1. Chemical composition of *Lavandula angustifolia* essential oil (%).

Compound	Retention time (min)	Percentage (%)
Alpha-pinene	6.072	0.169
Camphene	7.047	0.031
Beta-pinene	8.350	0.017
Sabinene	8.446	0.035
Beta-myrcene	9.791	0.871
Butanoic acid, butyl ester	10.895	0.212
3-octanone	12.050	3.974
Beta-ocimene	12.874	2.939
Ocimene	13.497	2.545
I-hexanol	14.582	0.060
3-octanol, acetate	17.347	0.234
3-octanol	18.049	0.464
Octen-1-ol, acetate	18.519	0.618
Neo-allo-ocimene	19.602	0.435
Butanoic acid, hexyl ester	22.142	1.076
Alcanfor/camphor	24.691	0.338
Linalool	26.469	37.023
Linalyl acetate	28.757	28.651
4-terpineol	29.864	3.191
Lavandulyl acetate	31.186	3.303
4-hexen-1-ol, 5-4methyl-2-(1-methylethenyl)	32.494	1.353
Borneol	33.821	0.530
Alpha-terpineol	34.203	1.410
Trans-caryophyllene	35.640	3.519
Neryl acetate	37.416	0.310
Beta-farnesene	38.463	5.090
Geraniol	41.026	0.653
Cumic alcohol	51.851	0.117
Oxtrane, tridecyl	79.284	0.722

essential oil on *E. coli* O157:H7 was determined via broth microdilution method. Briefly, 1 mL of the bacterial inoculum and 0.05 - 51.2 µL/mL of *L. angustifolia* essential oil were added to the test tubes containing 1 mL of Mueller Hinton Broth, and they were incubated at 37°C for 24 h. The final dilution factor without bacterial growth was accepted as the MIC value (Djenane *et al*., 2012).

Minimum bactericidal concentration (MBC)

The samples taken from the tubes with two lower and two upper values of the determined MIC value were inoculated onto the Mueller Hinton agar via the spread-plate method, and the plates were incubated at 37°C for 24 h. The tubes in which growth was not observed was determined to be MBC value.

Preparation of the meatball samples

Minced meat containing 90% meat and 10% fat was prepared. The bulk meatball sample was prepared so as to have a composition of 84% (w/w) minced meat, 2.0% (w/w) salt, 8.0% (w/w) breadcrumbs, 2.0% (w/w) red pepper, 0.1% (w/w) black pepper, 0.4% (w/w) cumin, 0.5% (w/w) garlic, and 3.0% (w/w) chopped onion. The bulk sample was divided into six equal parts. After homogenising, the

bulk sample were divided into 40 - 50 g each with ~5 cm in diameter. Six different groups of meatballs were prepared. While *E. coli* O157:H7 and essential oil were not added to group A (control group), group B contained the pathogen. MIC and 2-fold MIC doses of essential oil were added to pathogen-free groups C and D, respectively. MIC and 2-fold MIC doses of essential oil were added to groups E and F samples inoculated with *E. coli* O157:H7, respectively.

Meatball samples were wrapped with polyethylene film, and packaged in foam plates before being stored at 4°C until subsequent analysis. The microbiological and physiochemical evaluations were performed on days 0, 1, 3, 5, 7, 10, and 12. Sensory evaluation was performed on days 0, 3, 7, and 12.

Microbiological analysis

Meatball samples were analysed on the said days by mixing 10 g of sample with 90 mL of 0.1% (v/v) peptone water. The meatballs were then homogenised in a stomacher (IUL instruments, Barcelona, Spain). Then, the homogenates were diluted with 0.1% peptone water in ten-fold serial dilutions. Next, 1 mL of these dilutions were separately inoculated onto agar plates using the pour-plate technique. The total number of aerobic mesophilic bacteria (TAMB) was determined using plate count agar (PCA-Biolife, 4019352). The plates were incubated for 72 h at 30 ± 1°C (ISO, 2003). The potato dextrose agar (PDA; Biolife, 4019352) was used to count yeasts and moulds. The plates were incubated for 5 d at 22°C (ISO, 2008). The *E. coli* counts were determined using the Tryptone Bile X-Glucuronide medium (TBX-Oxoid CM0945). The plates were incubated for 4 h at 30°C, then 18 h at 44°C (ISO, 2006). The coliforms were determined using violet red bile agar (VRB-Merck 1.10275). The plates were incubated for 24 h at 30°C (ISO, 2006).

S. aureus was determined using the Baird Parker Agar supplemented with egg yolk tellurite emulsion. The plates were incubated at 37 ± 1°C for 48 h. Then, typical colonies (black, shiny, convex, with a narrow white entire margin and surrounded by clear zones extending 2 - 5 mm into the opaque medium) were determined, and coagulase test was performed. Five of the typical and atypical colonies were selected and incubated at 37°C for 24 h in Brain Heart Infusion (BHI) broth. Then, 0.1 mL sample was taken from each BHI broth culture, and mixed with 0.3 mL of sterile rabbit plasma. These tubes were incubated at 37°C. Initially, in the first 6 h, the tubes were controlled every 30 min, and the negative ones were incubated for up to 24 h (ISO, 1999b).

The isolation and identification of *E. coli* O157:H7 were performed following the method recommended by International Organization for Standardization (ISO, 2017). Briefly, 25 g of samples were mixed with 225 mL of novobiocin-containing modified TSB broth (mTSB broth). Then, the mixture was homogenised using a stomacher, and incubated at 37°C for 16 - 20 h. After incubation, 0.1 mL of samples were taken from this homogenate and inoculated into Cefixime Tellurite Selective Sorbitol MacConkey Agar (CT-SMAC) (Biolife, 40166952) followed by incubation at 42°C for 48 h. At the end of the incubation, sorbitol-negative colonies were inoculated to Fluorocult VRB Agar (VRB-MUG) (Biolife, 4021862), and incubated at 42°C for 48 h. The plates were checked whether or not they fluoresce using a UV hand lamp at 366 nm. Those that fluoresce were considered as positive for β-glucuronidase. The biochemical tests, namely the β-glucuronidase, Indole, Methyl Red, Voges-Proskauer, Citrate, Lysine Decarboxylase, and Triple Sugar Iron were applied to the formed colonies, and identified by applying *E. coli* O157:H7 Rapid latex agglutination test (ISO, 2001).

At the end of the incubation period under aerobic conditions, the plates with 30 - 300 colonies were enumerated (Harrigan, 1998), and the results were expressed as log CFU/g.

Physicochemical analysis

The pH value of meatball samples was determined using a digital pH meter (704 pH Meter, Metrohm) (ISO, 1999a). The salt content was determined using the Mohr's technique (AOAC, 1990). The dry matter was determined following the ISO 1442 method (ISO, 1997). The water activity was determined following the method specified by the Association of Official Agricultural Chemists (AOAC, 2019). Approximately, 15 g of each sample was taken and placed in the sample cabinet of the Testo 645 (Germany) device.

Lipid oxidation

The thiobarbituric acid reactive substances (TBARS) values were determined following the method proposed by Djenane *et al*. (2012) with slight modifications. Briefly, 10 g of meatball samples was homogenised with 20 mL of 10% (v/v) trichloroacetic

acid. The samples were centrifuged at 2,300 *g* for 30 min at 5°C. After filtering the homogenate through Whatman No:1 qualitative filter paper, 2 mL of the filtrate was added to 2 mL of TBA (20 mM) (Sigma-Aldrich). The mixture was then vortexed and incubated at 97°C for 20 min in boiling water to facilitate the development of pink colour. After the samples were cooled in cold water, absorbance was measured at 532 nm using a UV-visible spectrophotometer. Distilled water served as blank. The results were expressed as the average of three replicates per sample as mg malaondialdehyde/kg meatball.

Sensory analysis

The randomly selected 10 panellists who participated in the sensory evaluation of meatball samples were 25 - 45 years old from male and female postgraduate students at Faculty of Veterinary Medicine, Burdur Mehmet Akif Ersoy University, and staff members. The panellists were asked not to eat or drink anything at least 1 h prior to the study. To cleanse the palate after each sensory test, the panellists were permitted to drink water and to eat breadcrumbs. In order to eliminate the risk of interference, the panellists who participated in the sensory evaluation were placed in an odour-free room illuminated with white light. On each day of the study, the meatballs in groups A, C, and D (did not contain *E. coli* O157:H7, weighed 40 - 50 g, and 5 cm in diameter) were fried (175°C, 10 min) and served in white plastic plates. The panellists were asked to rate the organoleptic properties (colour, odour, taste, texture, and general acceptability) of the meatballs using a 9-point intensity scale (1 extremely bad, 5 average, 9 extremely good) (Smaoui *et al*., 2016).

Statistical analysis

All analyses were performed in triplicates. A total of six different meatball groups (groups A, B, C, D, E, and F) which contained *L. angustifolia* essential oil at different concentrations, and that did not contain essential oil and *E. coli* O157: H7 were examined for microbial and physicochemical properties throughout storage (days 0, 1, 3, 5, 7, 10, and 12). The homemade meatball samples were also evaluated by the panellists regarding their sensory properties on four different days of the storage period (days 0, 3, 7, and 12). The results were subjected to one-way analysis of variance (ANOVA) using the Statistical Package for the Social Sciences (SPSS) (Version 25.0; SPSS, Chicago, IL, USA). The statistical procedure, Tukey's test, was used to determine significant differences among the mean values, and those at a level of $p < 0.05$ were considered statistically significant. The results were expressed as mean ± standard deviation (SD).

Results and discussion

Composition of L. angustifolia essential oil

The essential oil of *L. angustifolia* consisted of 29 components such as linalool (37.02%), linalyl acetate (28.65%), beta-farnesene (5.09%), 3-octanone (3.97%), trans-caryophyllene (3.52%), and lavandulyl acetate (3.30%) as presented in Table 1.

Much work has been published on the chemical composition of *L. angustifolia* essential oil. It was observed that the amounts of linalool, linalyl acetate, and lavandulyl acetate in *L. angustifolia* essential oil differed from those reported in other studies (Djenane *et al*., 2012). The amounts of phenolic compounds in essential oil vary depending on the season, location, and processing factors (Martínez-Graciá *et al*., 2015). It was also observed that different parts of the plant contained phenolic compounds at different concentrations (Pombal *et al*., 2016).

Antioxidant activity (DPPH)

Vast number of studies published on the antioxidant effects of essential oils have reported that some components of the essential oils, especially phenols, can prevent or delay the adverse effects of free radicals (Rojano *et al*., 2008). In the present work, the essential oil used as food additive had positive effects on the sensorial properties of the meatballs, and it was predicted that it would also have positive impact on human health and on shelf-life of the product as they increased antioxidant stability in foods.

The inhibition (%) value of ascorbic acid and *L. angustifolia* essential oil were measured at 517 nm using a UV-visible spectrophotometer, and found to be 96.27 and 78.13%, respectively. The IC_{50} values of ascorbic acid and *L. angustifolia* essential oil were 5.2 and 94.82 μg/mL, respectively.

The results of the antioxidant studies performed on *L. angustifolia* were variable due to use of different solvents, extraction methods, concentrations of essential oil components, and cultivation conditions of lavender (Andrys *et al*., 2017). The antioxidant activity of lavender essential

oil is related to the bioactive compounds such as linalool and linalyl acetate in its composition. Kıvrak (2018) examined eight different *L. angustifolia* species grown in Anatolia, and determined the IC_{50} values to range from 91.56 to 105.08 μg/mL. A similar result (IC_{50} = 96.67 μg/mL) was reported in the study by Spiridon *et al*. (2011). In another study, the antioxidant properties of five different *Lavandula* spp. were investigated *in vitro.* It was found that while the IC_{50} values of *L. angustifolia* ranged from 95.60 to 110.36 μg/mL, the IC_{50} value of *L. hybrida* was 73.53 μg/mL (Robu *et al*., 2012).

Antibacterial activity of Lavandula angustifolia essential oil

The antibacterial properties of *L. angustifolia* essential oil against *E. coli* O157:H7 was evaluated by determining inhibition zones, MIC, and MBC. Dimethyl sulfoxide (DMSO) as negative control yielded no inhibition zone which indicated the lack of antimicrobial activity (Aneja and Joshi, 2010). Gentamicin as positive control yielded 18 ± 1.29 mm inhibition zone of *E. coli* O157:H7. The diameter of the inhibition zone for *L. angustifolia* essential oil was 12 ± 2.18 mm, which is similar to the results reported by Djenane *et al*. (2012). The MIC and MBC values were 6.4 and 12.8 μL/mL, respectively. This is in line with the findings reported by Djenane *et al*. (2012).

Microbiological changes in meatballs

The total aerobic mesophilic bacteria (TAMB) are important indicators providing information on food hygiene during production and storage. It was found that except for the first and last days of analysis ($p > 0.05$), as compared to the control groups (group A and B) ($p < 0.05$), the meatball groups containing *L. angustifolia* essential oil had a lower amount of TAMB. TAMB counts were higher for groups B, E, and F as compared to the groups that did not contain *E. coli* O157:H7. These results could have been due to the fact that PCA is not a selective medium, and many types of bacteria such as *Bacillus* spp., *E. coli*, and *S. aureus* can grow on it (Lis-Balchin, 2002). Some studies have reported that lavender at different concentrations is effective against many bacterial and fungal species (Lis-Balchin, 2002; Gómez-Estaca *et al*., 2010; Djenane *et al*., 2012). The data obtained revealed that changes in MIC value did not cause significant difference on the TAMB levels.

The yeast and mould counts are given in Table 2. Except the third and fifth days of analysis ($p > 0.05$), among all the groups, the yeast and mould counts in the groups containing *L. angustifolia* essential oil were found to be lower ($p < 0.05$). It has been reported that *L. angustifolia* essential oil and oil vapour showed antifungal activity especially against *Aspergillus niger* and *A. fumigatus* (Cavanagh and Wilkinson, 2002). The data obtained showed that EO had antifungal effect. However, this effect did not appear to be concentration-dependent.

Staphylococcus aureus is considered as one of most common pathogens causing foodborne diseases (Balaban and Rasooly, 2000). The coagulase test was applied to the colonies obtained from Baird Parker Agar (supplemented with egg yolk tellurite emulsion), but none gave positive results. Throughout the storage period, the counts of *Staphylococcus* spp. in group B was higher than that of group F ($p < 0.05$). Comparing groups A, C, and D, it was found that the counts of microorganism in group C and D were lower than that of the group A ($p < 0.05$), except for the first and tenth days of storage ($p > 0.05$). It was observed that inhibition was not concentration-dependent ($p < 0.05$). More effective results were observed only on the twelfth day of storage in groups D and F which contained *L. angustifolia* essential oil at higher concentrations. In groups A and B, the amount of *Staphylococcus* spp. was 5.36 and 5.53 log CFU/g, respectively. In groups D and F, ~2 log decrease was observed (3.56 and 3.73 log CFU/g) ($p < 0.05$). Similar results were reported in other studies, in which essential oil was more effective on *Staphylococcus* spp. (Djenane *et al*., 2012).

The inoculated samples were evaluated in terms of *E. coli* counts. It was observed that *E. coli* counts were lower in the samples containing *L. angustifolia* essential oil as compared to group B ($p < 0.05$). At the beginning of storage, *E. coli* counts in groups B, E, and F were 7.26, 6.05, and 5.91 log CFU/g respectively, and on day 12, the counts decreased to 6.66, 5.78, and 5.66 log CFU/g, respectively. The results showed that EO inhibited the growth of *E. coli* ($p < 0.05$). It was found out that this effect depended on the concentration of EO ($p < 0.05$), except for day 3 ($p > 0.05$).

On the first and last days of the analysis, a significant decrease was observed in the coliform counts in all groups ($p < 0.05$). When meatballs containing *L. angustifolia* essential oil at the same

Table 2. Effect of *Lavandula angustifolia* essential oil on the microbiological quality of the meatballs (log CFU/g).

	Day of storage at 4°C						
	0	**1**	**3**	**5**	**7**	**10**	**12**
	TAMB						
A	7.80 ± 0.55^{Bb}	8.08 ± 0.57^{ABCb}	7.98 ± 0.02^{Aab}	7.83 ± 0.02^{Ab}	7.74 ± 0.06^{Ab}	8.14 ± 0.02^{Aab}	8.51 ± 0.02^{Aa}
B	7.80 ± 0.05^{ABab}	8.23 ± 0.02^{Aa}	7.67 ± 0.14^{Bb}	7.89 ± 0.05^{Aab}	7.61 ± 0.04^{Ab}	7.69 ± 0.07^{Aab}	8.14 ± 0.61^{Aab}
C	7.93 ± 0.06^{ABa}	7.51 ± 0.06^{BCbc}	7.33 ± 0.02^{Cc}	6.62 ± 0.04^{Dd}	6.67 ± 0.07^{Dd}	7.90 ± 0.03^{Bab}	7.88 ± 0.57^{Aabc}
D	7.55 ± 0.07^{ABa}	7.31 ± 0.02^{Cb}	7.02 ± 0.04^{Dc}	6.62 ± 0.13^{Dd}	6.58 ± 0.05^{Dd}	7.41 ± 0.02^{Cb}	7.61 ± 0.04^{Aa}
E	7.91 ± 0.05^{Aabc}	8.13 ± 0.04^{ABa}	7.96 ± 0.02^{Aab}	7.62 ± 0.08^{Bbc}	7.48 ± 0.05^{Bc}	7.87 ± 0.06^{Babc}	8.08 ± 0.63^{Aa}
F	7.74 ± 0.06^{ABa}	7.48 ± 0.15^{BCb}	7.60 ± 0.09^{Bb}	7.17 ± 0.02^{Cc}	7.01 ± 0.03^{Cd}	7.24 ± 0.03^{Dc}	7.81 ± 0.08^{Aa}
	S. aureus						
A	4.20 ± 0.03^{Ab}	4.04 ± 0.01^{Ab}	4.26 ± 0.01^{Ab}	5.19 ± 0.01^{Aab}	5.08 ± 0.01^{Aa}	5.06 ± 0.01^{Ba}	5.36 ± 0.01^{Aa}
B	4.01 ± 0.03^{Cd}	4.04 ± 0.02^{ABd}	4.04 ± 0.03^{Cd}	5.03 ± 0.04^{Bc}	5.06 ± 0.05^{Ac}	5.29 ± 0.03^{Ab}	5.53 ± 0.11^{Aa}
C	4.10 ± 0.05^{BCc}	3.98 ± 0.04^{ABCd}	4.06 ± 0.06^{Ccd}	4.59 ± 0.08^{Cb}	4.62 ± 0.09^{Bb}	5.12 ± 0.01^{Ba}	3.99 ± 0.02^{Bcd}
D	4.10 ± 0.04^{BCb}	3.86 ± 0.07^{CDcd}	3.99 ± 0.03^{CDbc}	4.68 ± 0.15^{Ca}	3.61 ± 0.07^{Def}	3.73 ± 0.04^{Dde}	3.56 ± 0.07^{Cf}
E	4.16 ± 0.02^{ABc}	3.91 ± 0.01^{BCDd}	4.15 ± 0.02^{Bc}	4.94 ± 0.03^{Bb}	5.10 ± 0.04^{Aa}	5.14 ± 0.05^{Ba}	3.92 ± 0.06^{Bd}
F	4.19 ± 0.03^{ABa}	3.81 ± 0.09^{Dc}	3.94 ± 0.03^{Db}	3.80 ± 0.03^{Dc}	3.94 ± 0.02^{Cb}	3.98 ± 0.06^{Cb}	3.73 ± 0.07^{Cc}
	Yeast and mould						
A	6.92 ± 0.05^{ABd}	6.87 ± 0.04^{Ade}	6.82 ± 0.03^{Ce}	7.18 ± 0.02^{BCc}	8.27 ± 0.02^{Ab}	8.32 ± 0.03^{Bab}	8.37 ± 0.01^{Ba}
B	7.01 ± 0.03^{Ae}	6.94 ± 0.03^{Ae}	7.43 ± 0.02^{Ad}	7.72 ± 0.09^{Ac}	7.88 ± 0.07^{Bb}	8.43 ± 0.03^{Aa}	8.45 ± 0.02^{Aa}
C	5.87 ± 0.06^{Cd}	5.78 ± 0.02^{Dd}	5.63 ± 0.05^{Db}	7.12 ± 0.02^{Ca}	6.84 ± 0.04^{Cc}	6.87 ± 0.03^{Dbc}	6.95 ± 0.02^{Db}
D	5.97 ± 0.05^{Cd}	5.81 ± 0.11^{De}	5.48 ± 1.04^{Ef}	6.16 ± 0.04^{Dc}	6.80 ± 0.06^{CDb}	6.85 ± 0.02^{Dab}	6.95 ± 0.01^{Da}
E	6.86 ± 0.05^{Bc}	6.45 ± 0.07^{Bd}	6.92 ± 0.04^{Bc}	7.29 ± 0.03^{Ba}	6.86 ± 0.05^{Cc}	6.98 ± 0.04^{Cbc}	7.05 ± 0.04^{Cb}
F	6.88 ± 0.05^{ABc}	6.19 ± 0.02^{Ce}	6.91 ± 0.04^{BCbc}	7.06 ± 0.04^{Ca}	6.66 ± 0.07^{Dd}	7.03 ± 0.05^{Cab}	7.10 ± 0.02^{Ca}
	Coliform						
A	6.10 ± 0.03^{Ec}	7.17 ± 0.04^{Ba}	6.37 ± 0.05^{Bb}	6.15 ± 0.01^{Dc}	5.84 ± 0.02^{Dd}	5.63 ± 0.02^{De}	5.57 ± 0.02^{Ce}
B	7.16 ± 0.01^{Bb}	8.12 ± 0.01^{Aa}	7.19 ± 0.03^{Ab}	6.79 ± 0.10^{Ad}	7.12 ± 0.01^{Ab}	6.70 ± 0.08^{Ad}	7.00 ± 0.01^{Ac}
C	6.95 ± 0.03^{Ca}	6.80 ± 0.09^{Cb}	5.89 ± 0.03^{Cd}	6.31 ± 0.10^{CDc}	5.21 ± 0.03^{Ee}	5.01 ± 0.02^{Ef}	4.75 ± 0.06^{Dg}
D	6.65 ± 0.11^{Da}	6.34 ± 0.01^{Db}	5.58 ± 0.13^{Dc}	5.66 ± 0.06^{Ec}	4.63 ± 0.05^{Fde}	4.55 ± 0.05^{Fe}	4.72 ± 0.09^{Dd}
E	7.65 ± 0.06^{Aa}	7.12 ± 0.04^{Bc}	7.22 ± 0.03^{Ab}	6.45 ± 0.01^{Bd}	6.26 ± 0.01^{Be}	6.14 ± 0.02^{Bf}	5.92 ± 0.05^{Bg}
F	6.96 ± 0.48^{Ca}	6.83 ± 0.56^{Cb}	6.30 ± 0.63^{Bc}	6.14 ± 0.36^{Dd}	6.13 ± 0.81^{Cd}	5.99 ± 0.74^{Ce}	5.81 ± 0.80^{Bf}
	E. coli						
B	7.26 ± 0.02^{Ab}	7.46 ± 0.01^{Aa}	7.15 ± 0.01^{Ac}	6.68 ± 0.06^{Af}	6.70 ± 0.05^{Ade}	6.76 ± 0.03^{Ad}	6.66 ± 0.04^{Af}
E	6.05 ± 0.04^{Bc}	6.32 ± 0.01^{Ba}	5.69 ± 0.07^{Ce}	6.15 ± 0.02^{Bb}	6.13 ± 0.02^{Bbc}	5.84 ± 0.07^{Bd}	5.78 ± 0.04^{Bd}
F	5.91 ± 0.07^{Cb}	6.13 ± 0.59^{Ca}	5.87 ± 0.65^{Bb}	5.76 ± 0.39^{Cc}	5.94 ± 0.33^{Cb}	5.60 ± 0.51^{Cd}	5.66 ± 0.45^{Cd}
	***E. coli* O157:H7**						
B	8.91 ± 0.14^{Ab}	8.36 ± 0.03^{Ad}	7.82 ± 0.02^{Ae}	7.34 ± 0.02^{Af}	7.41 ± 0.01^{Af}	8.70 ± 0.04^{Ac}	9.14 ± 0.02^{Aa}
E	7.31 ± 0.07^{Ba}	7.37 ± 0.09^{Ca}	6.81 ± 0.45^{Cb}	5.33 ± 0.03^{Bc}	4.25 ± 0.03^{Be}	4.85 ± 0.07^{Bd}	4.33 ± 0.03^{Be}
F	6.98 ± 0.03^{Cb}	7.51 ± 0.05^{Ba}	6.16 ± 0.45^{Bc}	5.07 ± 0.03^{Cd}	3.56 ± 0.06^{Cg}	4.61 ± 0.06^{Ce}	4.18 ± 0.02^{Cf}

Values are expressed as mean ± standard deviation (SD). Means followed by different uppercase superscripts in the same column are significantly different ($p < 0.05$). Means followed by different lowercase superscripts in the same row are significantly different ($p < 0.05$). A = control, B = *E. coli* O157:H7, C = *L. angustifolia* essential oil 6.4 µL/mL, D = *L. angustifolia* essential oil 12.8 µL/mL, E = *E. coli* O157:H7 and *L. angustifolia* essential oil 6.4 µL/mL, F = *E. coli* O157:H7 and *L. angustifolia* essential oil 12.8 µL/mL.

concentration were compared, it was observed that the amount of coliforms in groups that contained *E. coli* O157:H7 was higher than that of the group which did not contain *L. angustifolia* essential oil ($p < 0.05$). When *L. angustifolia* essential oil was added at a concentration of two-fold of the MIC value, the highest inhibitory effect was observed against coliforms ($p < 0.05$).

E. coli O157:H7 inhibition was evaluated in the inoculated groups. The inhibition effect of *L. angustifolia* essential oil on *E. coli* O157:H7 is shown in Table 2. In group B, on the first day of analysis, *E. coli* O157:H7 was found to be 8.91 log CFU/g, and increased to 9.14 log CFU/g on day 12. In groups E and F, the number of microorganisms continued to decrease during storage. Aside from days 1 and 3 of analysis ($p > 0.05$), the highest inhibitory effect was observed for group F ($p < 0.05$). In a previous study, similar findings were reported for minced meat samples that contained essential oil at a concentration of two-fold of the MIC value (Djenane *et al*., 2012). Preliminary studies have revealed that *L. angustifolia* essential oil affected the log phase of *E. coli* O157:H7 by stopping the growth of bacteria and causing lysis (Sasaki *et al*., 2015).

Based on the biochemical and agglutination test results, all the colonies in group B which were suspected to be *E. coli* O157:H7, were confirmed as *E. coli* O157:H7. The colonies in groups E and F, which were suspected to be *E. coli* O157:H7, were confirmed as *E. coli* O157:H7 at 86% and 12%, respectively.

It was observed that *L. angustifolia* essential oil was effective on *E. coli* O157:H7 and coliforms in a dose-dependent manner. This was statistically meaningful ($p < 0.05$). The inhibition effect observed was not only due to the concentration of *L. angustifolia* essential oil but also due to the subspecies, as the synergistic effect among the various components of each subspecies has profound effect on microorganisms. Various subspecies of *Lavandula* have different essential oil composition depending on their genetic properties as well as the chemical and the physical features of the conditions they were grown in (Lis-Balchin *et al*., 1998).

Physicochemical changes in meatballs

Due to their high water activity, protein content, and approximately neutral pH, meat and meat products offer favourable conditions for bacterial growth (Angkititrakul *et al*., 2013). In the present work, throughout the storage period, no significant change was observed on water activity (a_w) among all groups.

The results of pH values were observed to be lower at the end of the storage period as compared to at the beginning ($p < 0.05$). Overall, the highest pH values were observed in the first two days of storage, and the changes in the pH ranged from 4.89 to 5.89. Especially at the end of the storage period, it was observed that the pH levels in the groups containing *L. angustifolia* essential oil at the level of 2×MIC were higher than that of the control groups ($p < 0.05$).

Dry matter and salt content of meatballs reached the highest values on day 5 ($p < 0.05$). The change in dry matter and salt content of the samples could not be related to the *L. angustifolia* essential oil concentration.

As can be seen in Table 3 and Figure 1, it can be assumed that except for the change in the pH level, the physicochemical changes depended neither on the presence nor the concentration of *L. angustifolia* essential oil. To the best of our knowledge, this is the first study in which these parameters were investigated.

Lipid oxidation level (TBARS)

Lipid oxidation is one of the major problems that deteriorates meat quality (Gray *et al*., 1996). The extent of lipolysis and oxidation lead to the increase in free fatty acids and flavour deterioration in meat products. The increase in TBARS value during the storage of meat products is related with the lipid oxidation (Turgut *et al*., 2017).

The TBARS values of the samples during storage at 4°C for 12 days are presented in Table 4. The initial concentration of TBARS in all samples were in the range of 0.49 to 0.67 mg MDA/kg. At the end of the storage, the TBARS values in groups A, C, and D reached 2.36, 1.24, and 1.14 mg MDA/kg, respectively. The TBARS of the control sample was significantly higher on day 1 to 12 than MIC and 2×MIC ($p < 0.05$). Therefore, it can be said that although the TBARS of both the control and treated meatballs increased over the storage period ($p < 0.05$), the concentration-dependent effect of *L. angustifolia* essential oil was observed to happen a rate lesser than the control group ($p < 0.05$). It was

Table 3. Effect of *Lavandula angustifolia* essential oil on the physicochemical properties of the meatballs.

	Day of storage at 4°C						
	0	**1**	**3**	**5**	**7**	**10**	**12**
	Water activity (a_w)						
A	0.98 ± 0.01	0.97 ± 0.01	0.95 ± 0.02	0.98 ± 0.01	0.96 ± 0.01	0.96 ± 0.01	0.97 ± 0.01
B	0.97 ± 0.01	0.98 ± 0.01	0.96 ± 0.02	0.97 ± 0.02	0.96 ± 0.02	0.96 ± 0.02	0.97 ± 0.01
C	0.97 ± 0.01	0.98 ± 0.01	0.97 ± 0.01	0.97 ± 0.01	0.96 ± 0.02	0.95 ± 0.02	0.97 ± 0.01
D	0.97 ± 0.01	0.97 ± 0.01	0.96 ± 0.02	0.97 ± 0.01	0.96 ± 0.02	0.96 ± 0.01	0.96 ± 0.03
E	0.97 ± 0.01	0.96 ± 0.01	0.96 ± 0.02	0.98 ± 0.01	0.97 ± 0.01	0.95 ± 0.03	0.96 ± 0.02
F	0.97 ± 0.01	0.96 ± 0.01	0.95 ± 0.01	0.98 ± 0.01	0.97 ± 0.02	0.97 ± 0.01	0.96 ± 0.03
	Dry matter (%)						
A	46.23 ± 0.01^{Cf}	49.05 ± 0.12^{Dc}	52.13 ± 0.14^{Db}	57.75 ± 0.10^{Da}	47.20 ± 0.25^{Ee}	47.18 ± 0.05^{Ce}	47.89 ± 0.11^{ABd}
B	50.76 ± 0.05^{Ae}	51.07 ± 0.18^{Ad}	53.59 ± 0.17^{Bb}	58.73 ± 0.01^{Ca}	51.58 ± 0.06^{Ac}	48.46 ± 0.06^{Af}	47.64 ± 0.01^{BCg}
C	47.75 ± 0.64^{Bd}	49.12 ± 0.14^{Dc}	52.41 ± 0.51^{CDb}	61.05 ± 0.11^{Aa}	48.01 ± 0.11^{Dd}	47.50 ± 0.01^{Bd}	46.22 ± 0.05^{Ee}
D	47.85 ± 0.16^{Be}	49.63 ± 0.11^{Cc}	52.02 ± 0.05^{Db}	60.24 ± 0.26^{Ba}	49.50 ± 0.21^{Bc}	48.54 ± 0.05^{Ad}	46.72 ± 0.12^{Df}
E	50.62 ± 0.59^{Ac}	51.29 ± 0.07^{Ac}	54.78 ± 0.15^{Ab}	57.08 ± 0.14^{Ea}	49.01 ± 0.20^{Cd}	48.36 ± 0.23^{Ade}	48.14 ± 0.16^{Ae}
F	48.35 ± 0.31^{Be}	50.59 ± 0.17^{Bc}	52.83 ± 0.14^{Cb}	57.66 ± 0.24^{Da}	49.60 ± 0.15^{Bd}	48.64 ± 0.01^{Ae}	47.50 ± 0.10^{Cf}
	pH						
A	5.79 ± 0.03^{Aa}	5.83 ± 0.04^{Aa}	5.61 ± 0.04^{Bb}	5.09 ± 0.03^{Db}	4.89 ± 0.03^{Ec}	5.08 ± 0.08^{Cb}	5.12 ± 0.04^{Bb}
B	5.87 ± 0.05^{Aa}	5.83 ± 0.02^{Aa}	5.68 ± 0.03^{ABb}	5.58 ± 0.05^{Ac}	5.40 ± 0.03^{ABd}	5.49 ± 0.02^{Acd}	5.15 ± 0.03^{Be}
C	5.80 ± 0.05^{Aa}	5.81 ± 0.02^{Aa}	5.70 ± 0.03^{Ab}	5.28 ± 0.04^{Ccd}	5.19 ± 0.03^{Dd}	5.30 ± 0.04^{Bc}	5.19 ± 0.02^{ABd}
D	5.80 ± 0.07^{Aa}	5.75 ± 0.03^{Aa}	5.71 ± 0.02^{Aa}	5.41 ± 0.03^{Bb}	5.22 ± 0.05^{CDc}	5.29 ± 0.03^{Bc}	5.27 ± 0.05^{Ac}
E	5.80 ± 0.07^{Aa}	5.83 ± 0.03^{Aa}	5.71 ± 0.03^{Ab}	5.59 ± 0.03^{Ac}	5.31 ± 0.04^{BCd}	5.35 ± 0.04^{Bd}	5.20 ± 0.03^{ABe}
F	5.89 ± 0.03^{Aa}	5.80 ± 0.07^{Aab}	5.70 ± 0.02^{Ab}	5.44 ± 0.06^{Bc}	5.41 ± 0.03^{Ac}	5.41 ± 0.04^{ABc}	5.27 ± 0.04^{Ad}
	Salt content (%)						
A	2.94 ± 0.01^{Df}	2.98 ± 0.01^{Ee}	2.98 ± 0.01^{Ee}	3.04 ± 0.01^{Da}	3.02 ± 0.01^{Bb}	3.02 ± 0.01^{Bc}	2.99 ± 0.01^{Bd}
B	3.09 ± 0.01^{Acd}	3.38 ± 0.01^{Ab}	3.40 ± 0.01^{Aa}	3.41 ± 0.01^{Aa}	3.09 ± 0.01^{Ac}	3.09 ± 0.01^{Acd}	3.08 ± 0.01^{Ad}
C	3.03 ± 0.01^{Bc}	3.05 ± 0.01^{Db}	3.05 ± 0.01^{Db}	3.08 ± 0.01^{Ca}	2.83 ± 0.01^{Dd}	2.83 ± 0.01^{Dd}	3.16 ± 0.58^{Dd}
D	2.98 ± 0.01^{Cc}	3.14 ± 0.01^{Bb}	3.15 ± 0.01^{Bb}	3.18 ± 0.01^{Ba}	2.87 ± 0.01^{Cd}	2.86 ± 0.01^{Cde}	2.85 ± 0.01^{Ce}
E	2.80 ± 0.01^{Fc}	3.07 ± 0.01^{Cb}	3.09 ± 0.01^{Cb}	3.17 ± 0.01^{Ba}	2.70 ± 0.01^{Fd}	2.70 ± 0.01^{Fd}	2.72 ± 0.01^{Fd}
F	2.87 ± 0.02^{Eb}	2.94 ± 0.01^{Fa}	2.94 ± 0.01^{Fa}	2.97 ± 0.03^{Ea}	2.76 ± 0.01^{Ec}	2.75 ± 0.01^{Ec}	2.77 ± 0.01^{Ec}

Values are expressed as mean ± standard deviation (SD). Means followed by different uppercase superscripts in the same column are significantly different ($p < 0.05$). Means followed by different lowercase superscripts in the same row are significantly different ($p < 0.05$). A = control, B = *E. coli* O157:H7, C = *L. angustifolia* essential oil 6.4 µL/mL, D = *L. angustifolia* essential oil 12.8 µL/mL, E = *E. coli* O157:H7 and *L. angustifolia* essential oil 6.4 µL/mL, F = *E. coli* O157:H7 and *L. angustifolia* essential oil 12.8 µL/mL.

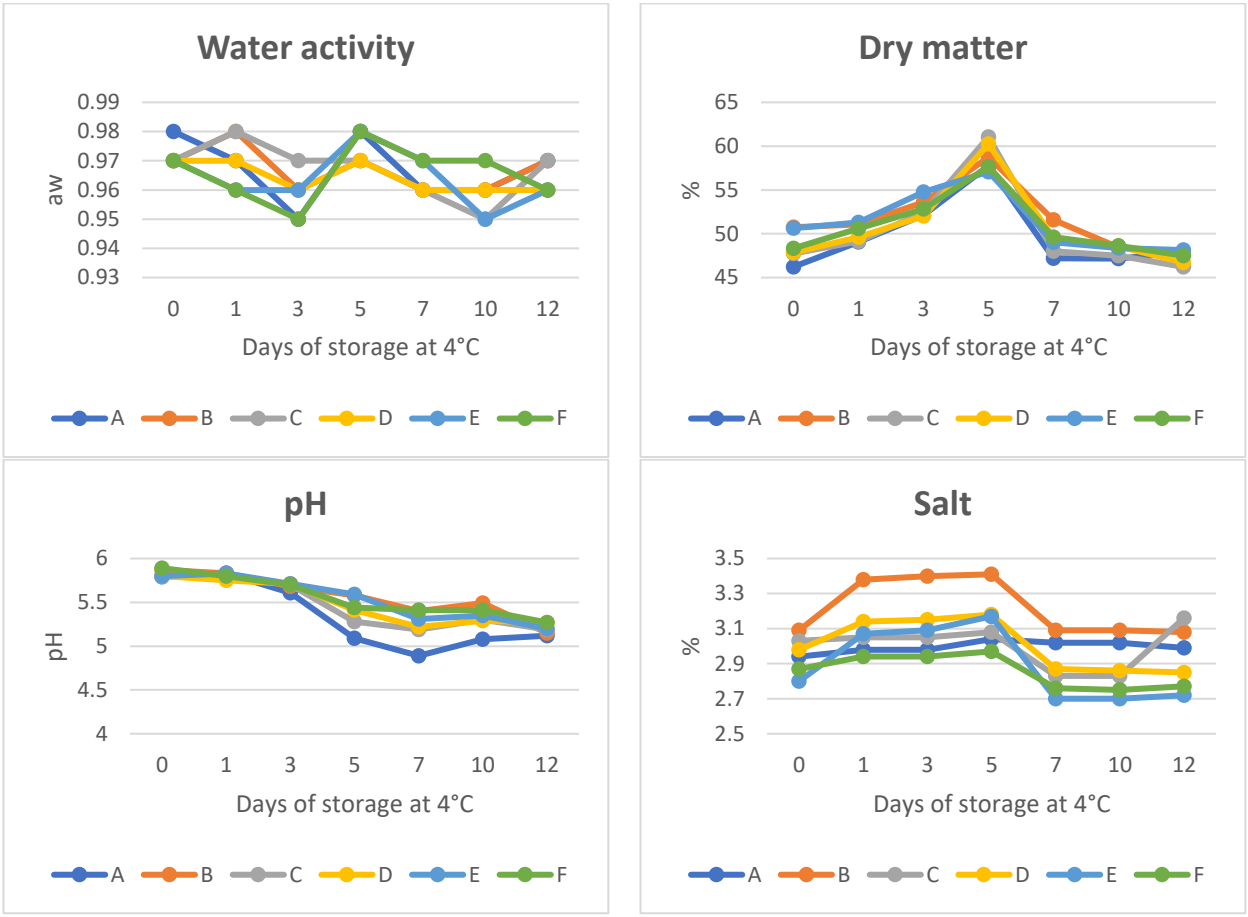

Figure 1. Effect of *Lavandula angustifolia* essential oil on the physicochemical properties of the meatballs. A = control, B = *E. coli* O157:H7, C = *L. angustifolia* essential oil 6.4 µL/mL, D = *L. angustifolia* essential oil 12.8 µL/mL, E = *E. coli* O157:H7 and *L. angustifolia* essential oil 6.4 µL/mL, F = *E. coli* O157:H7 and *L. angustifolia* essential oil 12.8 µL/mL.

Table 4. Effect of *Lavandula angustifolia* essential oil on the TBARS (mg MDA/kg) values of the meatballs.

	Day of storage at 4°C					
	1	**3**	**5**	**7**	**10**	**12**
A	0.67 ± 0.02^{Bf}	1.07 ± 0.05^{Be}	1.31 ± 0.01^{Cd}	1.75 ± 0.01^{Cc}	1.97 ± 0.02^{Cb}	2.36 ± 0.02^{Ca}
C	0.51 ± 0.02^{Af}	0.64 ± 0.02^{Ae}	0.81 ± 0.02^{Bd}	0.99 ± 0.03^{Bc}	1.16 ± 0.03^{Bb}	1.24 ± 0.02^{Ba}
D	0.49 ± 0.02^{Af}	0.60 ± 0.02^{Ae}	0.73 ± 0.03^{Ad}	0.81 ± 0.02^{Ac}	0.93 ± 0.04^{Ab}	1.14 ± 0.03^{Aa}

Values are expressed as mean ± standard deviation (SD). Means followed by different uppercase superscripts in the same column are significantly different ($p < 0.05$). Means followed by different lowercase superscripts in the same row are significantly different ($p < 0.05$). A = control, C = *L. angustifolia* essential oil 6.4 µL/mL, D = *L. angustifolia* essential oil 12.8 µL/mL.

determined that the addition of *L. angustifolia* essential oil could retard lipid oxidation during storage as compared to the control group. Similarly, Djenane *et al*. (2012) reported that the TBARS values for *L. angustifolia* essential oil added into minced beef samples were lower than the control group during storage period.

Depending on the lipid oxidation level, it can lead to either satisfactory or unsatisfactory flavour in meat products (Muriel *et al*., 2004). The value of 2 in TBARS is accepted as the limit point in meat; when this value is exceeded, the rancid flavour overpowers the meat flavour (Campo *et al*., 2006).

Sensorial changes in meatballs

It is well documented that the use of essential oils at high concentrations affected the organoleptic properties of foods (Martínez-Graciá *et al*., 2015). Meatball groups were analysed for their colour during the storage period. At the beginning of the storage period, group A had the best value ($p > 0.05$). However, groups containing *L. angustifolia* essential oil scored higher on the last day of the storage ($p < 0.05$) (Table 4).

On the first day of storage, the scores that groups A, C, and D got on texture were 8.27, 7.63, and 7.30, respectively. At the beginning, the control group got the higher scores than those groups containing *L. angustifolia* essential oil ($p < 0.05$). However, there was no significant difference between the groups on the last day on texture ($p > 0.05$).

At the beginning of the analysis, with respect to their aroma, group A was ranked as the most preferred group by the panellists. However, on the last day of the analysis, group C (6.70) was ranked as the most preferred group. It was also determined that the scent of lavender was reduced over the storage period since essential oil is volatile.

Regarding the taste of the meatballs, group A was the most preferred group (8.70). However, on the seventh day of storage, groups A (7.03) and C (6.97) received similar scores, and on the last day of storage, group C was the highest-ranking group (6.43) ($p < 0.05$).

On the first day, the general acceptability values of groups A and C were 8.57 points and 5.40 points, respectively. However, on day 12, both of the groups received good general acceptability scores (Table 5).

The results of the sensory analysis were promising since meatballs that contained *L. angustifolia* essential oil were not preferred due to the perception arising from an unknown flavour. However, in the later days of the analysis, lavender became more acceptable as the panellists got used to *L. angustifolia* essential oil in the meatballs.

Table 5. Sensory evaluation of the meatballs.

	Analysis day			
	0	**3**	**7**	**12**
	Colour			
A	8.50 ± 0.68^{Aa}	8.33 ± 1.27^{Aa}	7.90 ± 1.09^{Aa}	6.70 ± 1.60^{Bb}
C	8.20 ± 0.66^{ABa}	8.13 ± 1.20^{Aa}	7.83 ± 0.95^{Aa}	7.87 ± 1.07^{Aa}
D	8.03 ± 0.89^{Ba}	8.20 ± 1.06^{Aa}	8.10 ± 0.61^{Aa}	7.77 ± 0.90^{Aa}
	Texture			
A	8.27 ± 0.74^{Aab}	8.53 ± 0.68^{Aa}	7.73 ± 1.08^{Ab}	6.80 ± 1.49^{Ac}
C	7.63 ± 0.76^{Ba}	7.83 ± 1.21^{ABa}	7.57 ± 0.94^{Ab}	7.47 ± 1.20^{Aa}
D	7.30 ± 1.21^{Ba}	7.43 ± 1.65^{Ba}	7.53 ± 1.14^{Aa}	7.23 ± 1.33^{Aa}
	Odour			
A	8.57 ± 0.68^{Aa}	8.50 ± 0.68^{Aa}	7.20 ± 1.40^{Ab}	5.03 ± 1.40^{Bc}
C	5.53 ± 1.53^{Bb}	6.30 ± 1.29^{Bab}	7.03 ± 1.10^{Aa}	6.70 ± 1.39^{Aa}
D	3.73 ± 1.36^{Cb}	4.83 ± 1.29^{Ca}	5.73 ± 1.31^{Ba}	5.33 ± 1.49^{Ba}
	Taste			
A	8.70 ± 0.60^{Aa}	8.90 ± 0.31^{Aa}	7.03 ± 1.59^{Ab}	4.37 ± 1.77^{Bc}
C	4.80 ± 1.63^{Bc}	5.53 ± 1.55^{Bbc}	6.97 ± 1.10^{Aa}	6.43 ± 1.61^{Aab}
D	2.97 ± 1.22^{Cc}	3.93 ± 1.28^{Cb}	4.97 ± 1.25^{Ba}	4.70 ± 1.49^{Bab}
	General acceptability			
A	8.57 ± 0.68^{Aa}	8.50 ± 0.73^{Aa}	7.37 ± 1.10^{Ab}	5.27 ± 1.64^{ABc}
C	5.40 ± 1.52^{Bb}	6.23 ± 1.28^{Bab}	7.03 ± 0.85^{Aa}	6.30 ± 1.90^{Aab}
D	3.60 ± 1.10^{Cb}	4.53 ± 1.22^{Ca}	5.30 ± 1.09^{Ba}	4.93 ± 1.53^{Ba}

Values are expressed as mean ± standard deviation (SD). Means followed by different uppercase superscripts in the same column are significantly different ($p < 0.05$). Means followed by different lowercase superscripts in the same row are significantly different ($p < 0.05$). A = control, C = *L. angustifolia* essential oil 6.4 µL/mL, D = *L. angustifolia* essential oil 12.8 µL/mL.

Conclusion

Lavandula angustifolia essential oil had inhibitory effect on TAMB, coliforms, yeasts and moulds, *S. aureus*, *E. coli*, and especially on *E. coli* O157:H7 in the meatball samples. However, combinations with other food preservative ingredients might show better results. Aside from the interesting results obtained on its antimicrobial activity, the results of the sensory analysis were also promising. Over time, the groups that contained *Lavandula* became more preferable. Based on the habits and tolerance of consumers towards change, the meat products that contain *L. angustifolia* essential oil can be a contribution to the food industry with respect to food variety, health benefits, and solution to several food safety concerns. Since *L. angustifolia* essential oil has radical-scavenging properties, it can be used as antioxidant in meat products. *L. angustifolia* essential oil can also be used as an additive to retard the rancid flavour arising from lipid oxidation.

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