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Antimicrobial effect of *Thymus vulgaris* and *Rosmarinus officinalis* L. essential oils against Gram-positive bacteria inoculated in cooked turkey breasts stored under different temperatures

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Abstract: In order to simulate post-cooking contamination, the study evaluated the effects of storage temperature on the antibacterial activity of essential oils (EOs) of *Rosmarinus officinalis* L. (0.6%) and *Thymus vulgaris* (0.3%) added to cooked turkey breasts inoculated with *Staphylococcus aureus* (ATCC 25923), *Bacillus cereus* (ATCC 14579), and *Listeria innocua* (ATCC 33090). The turkey breasts were packaged aerobically and stored at 3 ± 1 °C for 120 h, 8 ± 1 °C for 120 h, and 3 ± 1 °C for 24 h, followed by 12 h at 25 ± 1 °C, and at 3 ± 1 °C until the end of storage (120 h). Sensory assessments and microbial analysis were performed at 0, 24, 36, 72, and 120 h of storage. A gas chromatography-mass spectrometry was used to analyse the components of the EOs; the major components in rosemary EO were 1,8-cineole (40.99%), α-pinene (13.40%), and campohr (10.92), while thyme EO was dominated by thymol (35.72%), *o*-cymene (20.65%), and γ-terpinene (8.81%). Thyme EO application led to a significant ($P < 0.05$) reduction of bacterial growth at each day of testing and was unaffected by abused storage temperature, while rosemary EO, when used at 0.6%, was found to be effective against *L. innocua* cells during regular and inadequate storage (3 ± 1 °C and 8 ± 1 °C). According to sensory analysis, samples containing thyme and rosemary EO received better scores than the control. These results demonstrate that thyme and rosemary EOs have significant antibacterial activity and can be recommended as a preservative except for post-contamination.

Keywords: essential oil emulsions; bacterial strains; cooked meat; storage temperature; post-cooking contamination

Foodborne illness resulting from consumption of food contaminated with pathogenic bacteria has been of vital concern to public health (Oussalah et al. 2007). Among the reported foodborne pathogens, *Staphylococcus aureus* (Yuan and Yuk 2018), *Bacillus cereus* (Ayari et al. 2016; López et al. 2015), and *Listeria monocytogenes* (Odedina et al. 2016; Yousefi et al. 2020) are a significant source of foodborne diseases. The European Food Safety Authority reported that the handling, preparation, and/or consumption of poultry meat can be a food-borne biological hazard due to several Gram-positive microorganisms (EFSA 2012). Additionally, it has been reported that some Gram-positive pathogens are capable of growing on

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refrigerated meat products, making refrigeration temperatures and post-process contamination a major concern for ready-to-eat meat products (Djenane 2011; Odedina et al. 2016).

To mitigate the risk of foodborne pathogen contaminations, innovative preservation methods, including plant-based bio-preservatives, have been suggested and extensively investigated (Barcenilla et al. 2022). Bio-preservatives or green chemicals, which are also called plant essential oils, are compounds that are naturally derived and known for their antiseptic properties. Currently, they are being considered as potential substitutes for chemical preservatives (Bensid et al. 2014; Thanissery et al. 2014). Essential oils (EOs) are volatile and lipophilic liquids extracted from a variety of plant organs through several techniques such as expression, fermentation, enfleurage, extraction, and the method of steam distillation and are known for their antimicrobial and antioxidant characteristics (Burt 2004; Valdivieso-Ugarte et al. 2019).

Among the EOs, thyme and rosemary EOs are natural additives that are used as a food ingredient in the Mediterranean region as aromatic and flavouring agents, well known for their antioxidative and antimicrobial properties (Djenane et al. 2011; Burt 2004). Moreover, the antibacterial activity of both thyme and rosemary EOs was investigated on a wide range of Gram-positive bacteria, including *Listeria monocytogenes*, *Staphylococcus aureus*, and *Bacillus cereus* (Govaris 2011; Guo et al. 2014; Ayari et al. 2016; Odedina et al. 2016; Yuan and Yuk 2018; Yousefi 2020).

Therefore, this work aims to characterise the impact of storage temperature on the sensory and antibacterial properties of essential oil emulsions of *Thymus vulgaris* and *Rosmarinus officinalis* against pathogenic strains (*S. aureus*, *B. cereus* and *L. innocua*) inoculated into cooked turkey breast cubes to simulate post-contamination.

MATERIAL AND METHODS

Bacterial strains and preparation of inocula. Lyophilised strains of *Bacillus cereus* (ATCC 14579), *Listeria innocua* (ATCC 33090), and *Staphylococcus aureus* (ATCC 25923) were acquired from MicroBiologics (USA). As explained below, each bacterial strain was first combined independently with the hydration fluid and then aseptically subcultured in selected culture media for 24 h at 37 ± 1 °C.

Fresh cultures were used to generate each microbial suspension, and colonies were individually injected into tubes containing 0.1% peptone water (TM MEDIA, India). Each culture's concentration was set at 0.5 McFarland (Liofilchem s.r.l., Italy).

Essential oils. Thyme and rosemary essential oils were purchased from Voshuiles' store (France). The EOs were certified by Ecocert FR-BIO 01 (France) and were obtained from Mediterranean biological culture by steam distillation and were then stored in sealed glass vials at 3 ± 1 °C until use.

Analysis of essential oil (GC-MS). The gas chromatography-mass spectrometry (GC-MS) analysis of the samples was carried out in the Technical Platform of PhysicoChemical Analysis (PTAPC-CRAPC, Algeria), using a Shimadzu GCMS QP2020 instrument, equipped with a fused Rxi®-5ms capillary column (Phase: Crossbond® 5% diphenyl/95% dimethyl polysiloxane). Its dimensions are 30 m × 0.25 mm and 0.25 μm film thickness. This column has a similar phase to the following columns: HP-1ms, HP-1msUI, DB-1ms, DB-5ms, DB-1msUI, Ultra-1, VF-1ms, ZB-1, and ZB-1ms and is considered also equivalent to USP G1, G2, and G38 phases. A volume of 250 μL of samples was injected in split mode (5 : 1). Injector and detector temperatures were maintained at 250 °C and 310 °C, respectively. The column temperature was programmed at 50 °C fixed for 2 min, then increased to 310 °C with an increase increment of 3 °C·min^{-1}, and then maintained at 310 °C for 2 min. The carrier gas used was helium (99.995% purity) with a flow rate of 1 mL·min^{-1}. The mass spectrometer conditions were as follows: ionisation voltage 70 eV, ion source temperature 200 °C, and electron ionisation mass spectra were acquired over the mass range of 45–600 m/z. The identity of the individual essential oil constituent was determined by comparing their mass spectra and time retention with those from Wiley 7N, NIST 02, and NIST 98 libraries. The percentage composition of the oils was calculated after normalising the areas of each component.

Emulsions preparation. Thyme and rosemary EOs emulsion was prepared using distilled water at 0.3% and 0.6%, respectively. 0.3 mL of Tween 80 (Merck, Germany) was added in order to ensure the miscibility of EOs in water. The solution was emulsified with a mechanical homogeniser for 2 min until a homogeneous mixture was obtained. The emulsion was used immediately after preparation for spraying the tested meat.

Preparation of turkey breasts. Turkey breasts, which were removed from the carcasses at 1 h post-mortem, were bought from a local poultry market. They were packaged in insulated polystyrene boxes

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and immediately transported to the laboratory under refrigerated conditions. After washing with cold sterile distilled water, the turkey breasts were left to drain on sterile steel wire mesh for 20 min in a refrigerator, cut, and weighted into portions of ≈10 g. Turkey breast samples were cooked in a microwave oven (LG 402TABN3P637, LG Electronics, Korea) at high power (700 W) for 5 min.

Treatments of samples and storage conditions. The cooked turkey breast samples were treated by spraying with 0.3% of thyme or 0.6% of rosemary essential oil emulsion and allowed to dry at room temperature for 5 min. The concentrations of plant extract and EOs yielding the best appearance and less presence of odour and colour in meat were chosen (Bensid et al. 2014).

The cooked meat samples were then inoculated separately by spreading 0.1 mL of inoculum (*S. aureus*, *L. innocua*, *B. cereus*, 10^6 CFU·mL^{-1}) across the surface of the samples so that the final concentration was approximately 10^4 CFU·g^{-1} of meat sample. Inoculated samples were kept in refrigerated temperatures for approximately 4–5 h for adhesion and absorption of inoculum.

Cooked turkey meat samples were randomly divided into nine separate groups: three groups (control 1, 2, and 3) were inoculated separately with *S. aureus*, *L. innocua,* and *B. cereus* strains but not treated by plant EO emulsions. Three other groups were treated with thyme essential oil emulsions, and inoculated separately with bacterial strains. Three other groups of samples were treated with rosemary essential oil emulsions and inoculated separately with bacterial strains. After inoculation and treatments, samples were packed separately in sterile polystyrene boxes with lids.

The tested meat samples were stored at different temperatures for 120 h, which is the expected shelf-life for cooked turkey meat; 3 ± 1 °C for 120 h (regular storage), 8 ± 1 °C for 120 h (poor storage, domestic refrigerator without thermometer), and 3 ± 1 °C for 24 h, followed by 12 h at 25 ± 1 °C and then at 3 ± 1 °C until the end of storage (abused storage). Groups were tested for *S. aureus*, *L. innocua,* and *B. cereus* counts after 0, 24, 36, 72, and 120 h of storage.

Sensory analysis. A panel of seven judges experienced in turkey evaluation was used for sensory analysis. Panellists were asked to evaluate odour, texture, appearance, and taste intensities of cooked samples. Acceptability of odour, texture, appearance, and taste was estimated using an acceptability scale ranging from 5 to 0, with 5 corresponding to a most liked sample and 0 corresponding to a least liked sample. A score of 3 was considered the lower limit of acceptability. Each evaluation was run in triplicate (n = 3), and every sample was evaluated individually.

Microbiological analysis. For each sampling day, 10 g of each sample was homogenised in 90 mL 0.1% buffered peptone water (TM Media, India) using a stomacher (Interscience, France). The homogenised samples were then ten-fold serially diluted and cultured on the following culture media: Baird Parker agar base supplemented with Egg Yolk Emulsion and Potassium Tellurite solution (TM Media) for *S. aureus*; Palcam agar base supplemented with a listeria selective supplement (TM Media) for *L. innocua*; and MYP agar supplemented with a Polymyxin B selective supplement and Egg Yolk Emulsion for *B. cereus* (TM Media). All plates were incubated at 37 ± 1 °C for 24 h and were examined visually for typical colony types and morphological characteristics associated with each medium. The bacterial count was expressed as $\log_{10}$ of the number of colony-forming units per gram of meat (log CFU·g^{-1}).

Statistical analysis. All experiments were carried out in triplicate, and the results are reported as the mean and standard deviation of these measurements. A one-way analysis of variance (ANOVA) was implemented, followed by Duncan's multiple range test using SPSS 19.0 for Windows (IBM SPSS Inc., USA). A P-value < 0.05 was used to evaluate the significant difference.

RESULTS AND DISCUSSION

GC-MS analysis. The GC/MS analysis of rosemary and thyme EOs revealed the presence of forty-eight and fifty-three constituents, accounting for 99.94% and 99.62% of the whole contents of the EOs, respectively. The major components in rosemary EO were 1,8-cineole (40.99%), α-pinene (13.40%), camphor (10.92%), β-pinene (5.34%), camphene (5.21%), and borneol (4.29%) (Table 1). Similar findings have been reported by Daferera et al. (2000, 2003); Pintore et al. (2002) and Ozogul et al. (2017). The components found in the highest concentrations in thyme EO were thymol (35.72%), *o*-cymene (20.65%) γ-terpinene (8.81%), carvacrol (8.28%), linalool (6.46%), and α-terpinene (2.63%) (Table 2). Previous studies have also shown major differences in the main constituents of thyme EO, where our results are also similar to those obtained by other authors (Özcan et Chalchat 2004; Lu et al. 2011). Lalonde et al. (2019) revealed the majority of which have excellent potential as meat-based antibacterial agents. The variability in composition and yields of the EOs

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Table 1. Chemical component of rosemary essential oil analysed by gas chromatography-mass spectrometry (GC/MS)

Name	Time of retention(min)	Composition (%)
Tricyclene	8.514	0.13
β-Phellandrene	8.700	0.17
α-Pinene	8.973	13.40
Camphene	9.523	5.21
β-Pinene	10.65	5.34
3-Octanone	11.04	0.05
Myrcene	11.26	1.58
α-Phellandrene	11.81	0.24
δ-3-Carene	12.07	0.29
α-Terpinene	12.35	0.53
o-Cymene	12.71	2.37
1,8-Cineole	13.12	40.99
(E)-β-Ocimene	13.78	0.04
γ-Terpinene	14.23	0.71
trans-4-Thujanol	14.59	0.04
α-Terpinolene	15.58	0.46
Linalool	16.10	1.09
Fenchol	16.72	0.11
Pinocarveol	17.89	0.04
Camphor	18.19	10.92
Dihydrocamphene carbinol	18.33	0.08
Borneol	19.18	4.29
Terpinen-4-ol	19.69	1.13
Cuminol	20.05	0.03
L-α-Terpineol	20.34	2.92
Myrtenol	20.58	0.04
Fenchyle acetate	24.67	0.49
Thymol	24.95	0.07
Carvacrol	25.37	1.24
α-Cubebene	27.51	0.02
Ylangene	28.45	0.11
Copaene	28.64	0.40
Methyleugenol	29.82	0.05
Caryophyllene	30.52	3.35
β-Calarene	30.88	0.05
Aromandendrene	31.30	0.07
(Z,Z,Z)-1,5,9,9-tetramethyl-1,4,7-cycloundecatriene	31.90	0.42
trans-Cadina-1(6),4-diene	32.72	0.01
γ-Cadinene	32.85	0.31
Muurola-4,9-diene	32.99	0.03
β-helmiscapene	33.24	0.03
10S,11S-Himachala-3(12),4-diene	33.59	0.10
α-Muurolene	33.81	0.11
β-Bisabolene	34.13	0.09
3,7(11)-Selinadiene	34.36	0.18
δ-Cadinene	34.73	0.49
α-Calacorene	35.48	0.02
Oxyde de caryophyllene	37.05	0.10

from the same plant can be dependent on conditions of growth (Theis and Lerdau 2003); harvesting time; variable ecological and geographical locations; the age of the plant; even different parts of the plant itself; and methods used for extraction (Kohiyama et al. 2015).

Microbiological assessments. Storage temperature yielded a significant difference ($P < 0.05$) between the treated groups all over the storage time and had an impact on the efficiency of EOs.

At 3 ± 1 °C, statistical analysis showed significant ($P < 0.05$) differences between the control and treatment groups where, *B. cereus*, *S. aureus*, and *L. innocua* values were significantly higher ($P < 0.05$) in control samples than in treated samples by the end of storage time (Table 3), which can be explained by the fact that the use of EOs has a significant effect in slowing down bacterial growth. The application of thyme EO decreased *B. cereus* and *S. aureus* strains' counts by 0.66 and 0.73 log CFU·g^{-1}, respectively, compared to the control groups, indicating that thyme EO was more effective than rosemary EO, especially at the end of the storage period. Thyme essential oil's chemical composition, which includes a high concentration of thymol (35.72%), is responsible for its antibacterial properties. Thymol affects energy production systems by influencing ATP-producing enzymes and altering ionic permeability, which permits ions to move across the aqueous extracellular environment (Hyldgaard et al. 2012). However, *L. innocua* values in samples treated with rosemary EO at 0.6% presented a different profile; the population of *L. innocua* in these samples was significantly decreased ($P < 0.05$) by 0.12 log CFU·g^{-1} after 72 h of storage at 3 ± 1 °C compared to the control group. The outcome highlights the remarkable impact of the species or strain on the various antimicrobial substances (Raybaudi et al. 2006). Moreover, the high content of 1,8-cineole in rosemary essential oil (40.99%) is responsible for its antibacterial properties. This

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Table 2. Chemical component of thyme essential oil analysed by gas chromatography-mass spectrometry (GC/MS)

Name	Time of retention(min)	Composition (%)
α-Phellandrene	8.696	0.84
α-Pinene	8.949	2.21
Camphene	9.516	0.66
β-Pinene	10.651	0.17
1-Octen-3-ol	10.726	0.20
Octan-3-one	11.070	0.09
Myrcene	11.271	2.44
β-Phellandrene	11.811	0.44
δ-Carene	12.072	0.15
α-Terpinene	12.359	2.63
o-Cymene	12.809	20.65
Linalyl butyrate	12.915	1.44
γ-Terpinen	14.298	8.81
Sabinene hydrate	14.595	0.32
trans-Linalool oxide	14.863	0.13
trans-$_D$-dihydrocarveol	15.590	0.54
Linalool	16.198	6.46
1-Acetyl-2-methyl-1-cyclopentene	17.171	0.08
trans-Pinocarveol	17.915	0.06
trans-Chrysanthemal	18.415	0.09
Bornyl formate	19.152	0.64
(-)-4-Terpineol	19.709	0.75
p-Cymen-8-ol	20.030	0.02
Cuminyl acetate	20.211	0.03
$_L$-α-Terpineol	20.310	0.16
trans-Dihydrocarvone	20.845	0.02
8,9-Dehydrothymol	21.486	0.02
Bornyl formate	22.057	0.03
α-Ocimene	22.240	0.01
cis-Geraniol	22.370	0.03
Isothymol methyl ether	22.776	0.50
Neral	23.003	0.02
Linalyl acetate	23.627	0.49
Benzyl alcohol	24.352	0.16
Thymol	25.317	35.72
Carvacrol	25.613	8.28
2-Oxabicyclo[2.2.2]octan-6-one, 1,3,3-trimethyl-	25.805	0.02
Eugenol	27.813	0.03
Copaene	28.654	0.04
Neryl acetate	28.957	0.01
α-Gurjunene	30.101	0.02
Caryophyllene	30.538	2.96
γ-Cadinene	30.851	0.02
(-)-α-Panasinsen	31.142	0.01
Benzenemethanol, 2-methoxy-α-methyl-	31.347	0.35
α-Humulene	31.912	0.07
10s,11s-Himachala-3(12),4-diene	32.206	0.02
γ-Muurolene	32.845	0.03
β-Copaene	33.030	0.03
Viridiflorene	33.613	0.09
α-Muurolene	33.810	0.02
γ-Cadinene	34.359	0.04
δ-Cadinene	34.725	0.11
(-)-Spathulenol	36.823	0.03
Caryophyllene oxide	37.043	0.07

component can harm cell membrane structure even at sub-inhibitory concentrations, according to a prior study (de Sousa et al. 2015).

At each testing day at 8 ± 1 °C, bacterial strains in both treated groups, were significantly affected by plant extract application and were higher ($P < 0.05$) in control than in treated samples until the end of storage time (Table 4). Additionally, thyme EO showed a higher anti-staphylococcal effect than rosemary at 8 ± 1 °C and increased its efficiency at this temperature compared to its effectiveness at 3 ± 1 °C. This finding supports that storage temperature was determinant on the growth of *S. aureus*. However, *L. innocua* bacteria count decreased by 0.41 log CFU·g^{-1} in samples treated with rosemary EO compared to thyme EO, while its effect is less effective during this storage temperature than during refrigerated storage. The volatility of the essential oil molecules may be the reason for the modest reduction in efficacy.

At abused storage (3 ± 1 °C for 24 h, followed by 12 h at 25 ± 1 °C and then at 3 ± 1 °C until the end of storage), the initially recorded population of 5.73 log CFU·g^{-1} of *B. cereus*, 5.20 log CFU·g^{-1} of *S. aureus,* and 5.22 log CFU·g^{-1} of *L. innocua* strains increased to approximately 6.07, 6.10, and 6.11 log CFU·g^{-1}, respectively, by the end of storage in untreated samples (Table 5). We presume this is due to abused storage temperature, which was frequently detected during one or more phases in the storage, during transpor-

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Table 3. Evolution of bacterial count in differently treated cooked turkey breasts stored at 3 ± 1 °C (mean ± SD, n = 3)

Sampling hour		H0	H24	H36	H72	H120
Bacillus cereus	C	5.73 ± 0.05^{Xb}	5.73 ± 0.11^{Yb}	5.79 ± 0.09^{Xb}	5.81 ± 0.03^{Xb}	6.00 ± 0.01^{Xa}
	R	5.73 ± 0.05^{Xa}	5.32 ± 0.24^{Yb}	5.14 ± 0.02^{Zb}	5.94 ± 0.01^{Xa}	5.88 ± 0.02^{Ya}
	T	5.73 ± 0.05^{Xa}	5.08 ± 0.11^{Yc}	5.29 ± 0.05^{Yb}	5.04 ± 0.08^{Zc}	5.07 ± 0.01^{Zc}
Staphylococcus aureus	C	5.20 ± 0.18^{Xb}	5.27 ± 0.50^{Xb}	5.55 ± 0.12^{Xab}	5.87 ± 0.28^{Xa}	5.91 ± 0.05^{Xb}
	R	5.20 ± 0.18^{Xa}	4.92 ± 0.32^{Xab}	4.74 ± 0.28^{Yb}	4.76 ± 0.04^{Yb}	4.71 ± 0.08^{Yb}
	T	5.20 ± 0.18^{Xab}	5.28 ± 0.73^{Xa}	4.56 ± 0.45^{Yab}	4.61 ± 0.02^{Yab}	4.47 ± 0.15^{Zb}
Listeria innocua	C	5.22 ± 0.12^{Xc}	5.48 ± 0.12^{Xb}	5.53 ± 0.15^{Xb}	5.58 ± 0.05^{Xb}	5.81 ± 0.01^{Xa}
	R	5.22 ± 0.12^{Xa}	4.78 ± 0.12^{Yb}	5.20 ± 0.08^{Ya}	5.10 ± 0.05^{Ya}	5.17 ± 0.02^{Ya}
	T	5.22 ± 0.12^{Xab}	5.00 ± 0.13^{Yb}	5.36 ± 0.02^{XYa}	5.37 ± 0.22^{Xa}	5.18 ± 0.17^{Yab}

$^{a–c}$different letters in the same row indicate a significant difference ($P < 0.05$) between sampling hours in the same temperatures for the same treatment; $^{X–Z}$different letters in the same column indicate significant differences ($P < 0.05$) between the different treatments in the same temperature on the same sampling hour; C – control; R – rosemary; T – thyme

Table 4. Evolution of bacterial count in differently treated cooked turkey breasts stored at 8 ± 1 °C (mean ± SD, n = 3)

Sampling hour		H0	H24	H36	H72	H120
Bacillus cereus	C	5.73 ± 0.05^{Xab}	5.73 ± 0.27^{Xc}	5.61 ± 0.22^{Xb}	5.82 ± 0.08^{Xab}	6.02 ± 0.01^{Xa}
	R	5.73 ± 0.05^{Xa}	5.03 ± 0.14^{Xc}	5.37 ± 0.07^{Xb}	5.55 ± 0.03^{Yab}	5.74 ± 0.20^{Ya}
	T	5.73 ± 0.05^{Xa}	5.08 ± 0.04^{Xb}	5.07 ± 0.08^{Yb}	4.78 ± 0.02^{Zc}	5.15 ± 0.01^{Zb}
Staphylococcus aureus	C	5.20 ± 0.18^{Xc}	5.29 ± 0.00^{Xbc}	5.44 ± 0.05^{Xab}	5.53 ± 0.12^{Xa}	5.55 ± 0.03^{Xbc}
	R	5.20 ± 0.18^{Xa}	4.94 ± 0.23^{Yb}	4.85 ± 0.05^{Yb}	5.24 ± 0.09^{Ya}	4.68 ± 0.03^{Yb}
	T	5.20 ± 0.18^{Xa}	4.72 ± 0.19^{Yb}	4.78 ± 0.10^{Yb}	4.66 ± 0.05^{Zb}	4.21 ± 0.16^{Zc}
Listeria innocua	C	5.22 ± 0.12^{Xab}	4.49 ± 0.35^{Yc}	4.99 ± 0.10^{Zb}	5.39 ± 0.33^{Xab}	5.60 ± 0.04^{Ya}
	R	5.22 ± 0.12^{Xb}	5.45 ± 0.31^{Xab}	5.58 ± 0.02^{Xa}	5.45 ± 0.09^{Xab}	5.19 ± 0.02^{Zb}
	T	5.22 ± 0.12^{Xc}	5.26 ± 0.24^{Xc}	5.39 ± 0.03^{Yc}	5.63 ± 0.05^{Xb}	5.99 ± 0.02^{Xa}

$^{a–c}$different letters in the same row indicate a significant difference ($P < 0.05$) between sampling hours in the same temperatures for the same treatment; $^{X–Z}$different letters in the same column indicate significant differences ($P < 0.05$) between the different treatments in the same temperature on the same sampling hour; C – control; R – rosemary; T – thyme

Table 5. Evolution of bacterial count in differently treated cooked turkey breasts stored at 3 ± 1 °C and temperature abuse (mean ± SD, n = 3)

Sampling hour		H0	H24	H36	H72	H120
Bacillus cereus	C	5.73 ± 0.05^{Xbc}	5.64 ± 0.17^{Xc}	6.09 ± 0.10^{Xa}	5.83 ± 0.04^{Xb}	6.07 ± 0.01^{Xa}
	R	5.73 ± 0.05^{Xb}	4.99 ± 0.25^{Yc}	5.70 ± 0.05^{Yb}	5.87 ± 0.07^{Xab}	5.97 ± 0.01^{Ya}
	T	5.73 ± 0.05^{Xa}	5.05 ± 0.17^{Yd}	5.40 ± 0.14^{Zb}	5.30 ± 0.07^{Ybc}	5.18 ± 0.02^{Zcd}
Staphylococcus aureus	C	5.20 ± 0.18^{Xb}	5.96 ± 0.00^{Xa}	6.08 ± 0.02^{Xa}	6.10 ± 0.06^{Xa}	6.12 ± 0.04^{Xa}
	R	5.20 ± 0.18^{Xb}	5.24 ± 0.18^{Yb}	5.60 ± 0.12^{Ya}	5.88 ± 0.07^{Ya}	5.08 ± 0.22^{Yb}
	T	5.20 ± 0.18^{Xbc}	5.10 ± 0.15^{Ybc}	5.52 ± 0.09^{Ya}	5.30 ± 0.02^{Zab}	5.04 ± 0.13^{Yc}
Listeria innocua	C	5.22 ± 0.12^{Xb}	5.37 ± 0.30^{Xb}	6.16 ± 0.01^{Xa}	6.03 ± 0.08^{Xa}	6.11 ± 0.02^{Ya}
	R	5.22 ± 0.12^{Xd}	5.07 ± 0.10^{XYe}	5.64 ± 0.07^{Zc}	5.94 ± 0.02^{XYb}	6.23 ± 0.02^{Xa}
	T	5.22 ± 0.12^{Xc}	4.92 ± 0.15^{Yd}	5.94 ± 0.05^{Ya}	5.88 ± 0.04^{Ya}	5.39 ± 0.01^{Zb}

$^{a–e}$different letters in the same row indicate a significant difference ($P < 0.05$) between sampling hours in the same temperatures for the same treatment; $^{X–Z}$different letters in the same column indicate significant differences ($P < 0.05$) between the different treatments in the same temperature on the same sampling hour; C – control; R – rosemary; T – thyme

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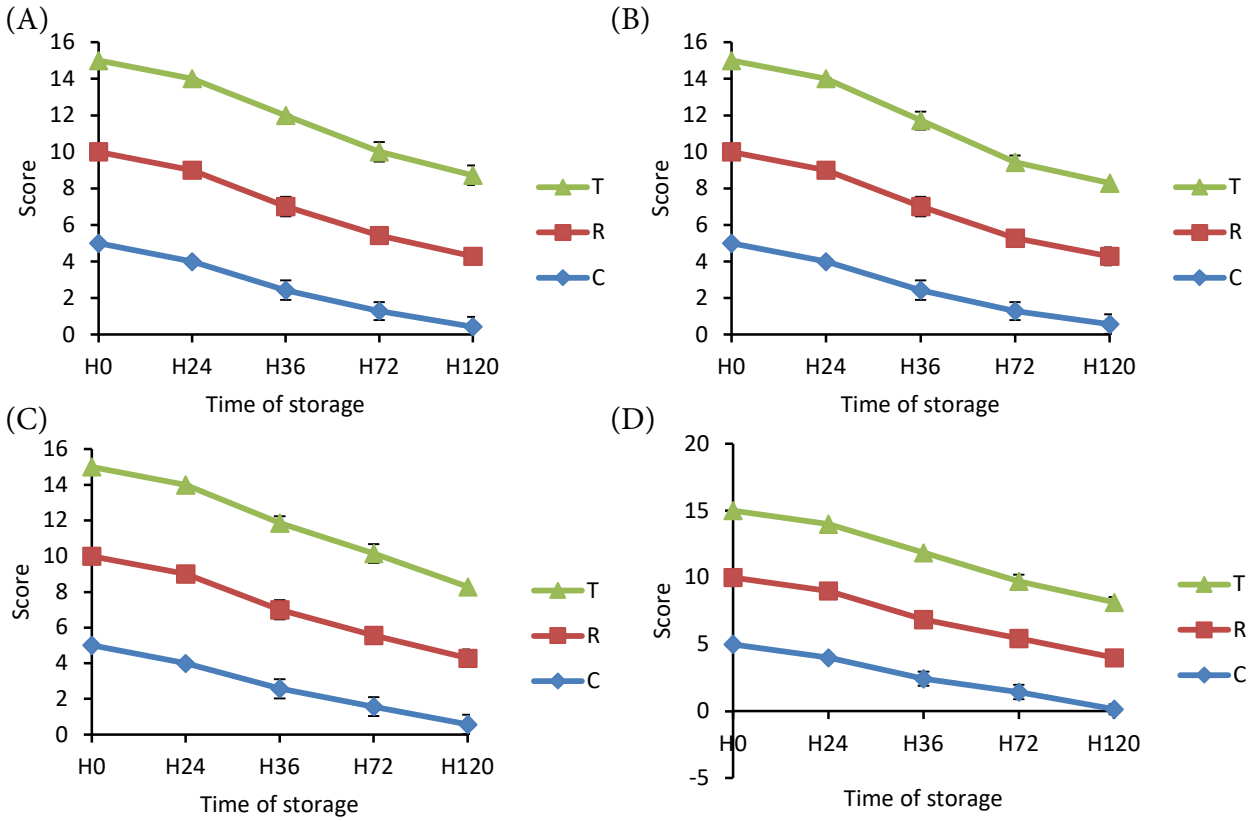

Figure 1. Sensory analysis of cooked turkey breasts treated with essential oil of thyme at 0.3% and essential oil of rosemary at 0.6% and stored at 3 °C for 120 h. Sensory attributes: (A) odour; (B) texture; (C) appearance; (D) taste intensities

C – control; R – rosemary; T – thyme

tation, or in domestic refrigerators (Djokhdem et al. 2022). Furthermore, the application of EOs yielded a significant difference ($P < 0.05$) between the treated groups all over the storage time, in which the application of thyme essential oil resulted in the lowest bacterial count values of the three strains during storage at excessive temperature, compared to rosemary essential oil, particularly towards the end of the storage period. This result may be explained by the fact that the bioactive components of thyme essential oil were also unaffected by the temperature conditions of abuse (Houicher et al. 2020).

According to the results of this study, the antibacterial activity of thyme essential oil was not influenced by storage conditions even after exposing cooked turkey breast cubes to an excessive temperature of 2 ± 1 °C. Hence, all strains showed their sensitivities to the components of thyme essential oil.

Sensory analysis. Throughout the storage period, significant differences ($P < 0.05$) were observed between the control and treated groups. However, significant differences ($P < 0.05$) were observed between the treated batches. The treated groups were highly preferred by the panellists due to their desirable flavour. A score of 3 was considered the lower limit of acceptability. The lowest acceptability score of 3 was obtained for odour, texture, appearance, and taste intensity after 36 h for the control group. We suggest that this is due to the increased microbial load, fat oxidation, protein degradation, and volatility of EO compounds. The limit of acceptability was reached only after 120 h of storage for the treated batches. However, samples containing thyme EO scored higher than samples containing rosemary EO (Figure 1). This could be explained by a high content of thymol, a bioactive phenolic compound that improves the organoleptic properties of cooked turkey breasts (Hyldgaard et al. 2012). Based on the sensory analysis of all samples, it can be concluded that 0.3% and 0.6% of thyme and rosemary EOs, respectively, could be used to preserve the organoleptic quality of cooked turkey breasts.

CONCLUSION

At abused storage, thyme EO application resulted in lower values of bacteria counts compared to rose-

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mary EO particularly toward the conclusion of the storage period. These results showed that the antimicrobial activity of thyme EO was unaffected by improper storage temperature, contrary to rosemary EO. In this study, it can be concluded that thyme and rosemary EOs has significant antibacterial activity and can be recommended as a preservative to increase the meat quality and safety and to improve organoleptic attributes for turkey breasts. The synergistic antibacterial actions of thyme and rosemary essential oils should be further investigated, especially in situations of excessive storage.

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