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Antimicrobial efficiency of rosemary, thyme and clove essential oils on the preservation of marinated chicken breasts (fillets)

Eficacia antimicrobiana de los aceites esenciales de romero, tomillo y clavo en la conservación de pechugas de pollo marinadas (filetes)

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ABSTRACT

Essential oils can be used as natural preservatives in the poultry meat industry. The aim of this research was to determine the effect of some essential oils on the microbial, physicochemical, and sensory properties of marinated chicken breast. For this purpose, rosemary, thyme, and clove essential oils were used at doses of 125 mg·kg^{-1} and 250 mg·kg^{-1} while marinating chicken breasts. After the marinated chicken breasts were divided into groups, they were stored in the refrigerator at 4°C. The results showed that 250 mg·kg^{-1} doses of essential oils, especially at 24 h, and rosemary had more inhibitory effects on some microbial (total mesophilic aerobic, total psychrophilic aerobic, and yeast–mold) parameters. However, the sensory groups with the addition of 125 mg·kg^{-1} were more accepted. Among these groups, the most acceptable group was the group that added 125 mg·kg^{-1} of thyme essential oil. The study shows that the use of thyme essential oil as an alternative to chemical preservatives may be beneficial both in extending the shelf life of marinated chicken breasts and in terms of consumer taste.

Key words: Essential oil compounds; marination; chicken breast; preservation; quality parameters

RESUMEN

Los aceites esenciales se pueden usar como conservantes naturales en la industria de la carne de aves de corral. El objetivo de esta investigación fue determinar el efecto de algunos aceites esenciales sobre las propiedades microbianas, fisicoquímicas y sensoriales de la pechuga de pollo marinada. Para este propósito, se usaron aceites esenciales de romero, tomillo y clavo en dosis de 125 mg·kg^{-1} y 250 mg·kg^{-1} mientras se marinaban las pechugas de pollo. Después de dividir las pechugas de pollo marinadas en grupos, se almacenaron en el refrigerador a 4°C. Los resultados mostraron que dosis de 250 mg·kg^{-1} de aceites esenciales, especialmente a las 24 horas, y romero tuvieron más efectos inhibidores sobre algunos parámetros microbianos (aeróbico mesófilo total, aeróbico psicrofílico total y moho de levadura). Sin embargo, los grupos sensoriales con la adición de 125 mg·kg^{-1} fueron más aceptados. Entre estos grupos, el grupo más aceptable fue el grupo que agregó 125 mg·kg^{-1} de aceite esencial de tomillo. El estudio muestra que el uso de aceite esencial de tomillo como alternativa a los conservantes químicos puede ser beneficioso tanto para extender la vida útil de las pechugas de pollo marinadas como en términos del gusto del consumidor.

Palabras clave: Compuestos oleosos esenciales; marinado; pechuga de pollo; conservación; parámetros de calidad

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INTRODUCTION

Nowadays, nutritionists have stated that an adult need to obtain 1/3 of the daily protein from animal-source foods. The reasons why the poultry meat being of great importance among animal-source foods is preferred may include having thinner fibers than butchery animal meat, low connective tissue and fat ratio, being prepared in a short time, easy serviceability, containing almost all of the amino acids that are essential for human nutrition, a quality protein structure, less energy and calories, easily digestibility, rich in B group vitamins and iron, lower prices than red meat, lower cost of production and high nutritional value [1, 2, 3, 4].

Contamination of chicken (*Gallus gallus domesticus*) meat and products with microorganisms is unavoidable. The microbiological flora in the "chicken meat" is composed of different microorganisms some of them are pathogens (like *Salmonella* and *Campylobacter*) for human, while others are spoilage bacteria (such as *Pseudomonas*, *Enterobacteria*). The level of microorganisms varies according to the applied hygienic conditions, the storage time and temperature of the products during the process from farm to dining table [5, 6, 7]. Food infections and poisoning may occur when sanitation and hygienic conditions are not followed during chopping, packaging, distribution and storage in the slaughterhouse chain [5, 8, 9]. Due to these reasons, it is very important to extend the shelf life of chicken meat and products by minimizing microbial activities without compromising their quality characteristics while on the shelves for consumer purchase. There are several preservation methods used for this purpose. Examples of advanced processing technologies are emulsion technology, coating technology, canning technology, curing and marination processes [10].

The word 'marination' derives from the Italian word 'marinare'. The word 'marination', which has been used since 1600s, means the preservation of meat and meat products by curing them with salt [11]. Today, the term 'marination' is stated as a method applied to has a remarkable effect on the evolution of the microbial growth and positively affects the sensory quality (taste, texture, juiciness, flavor) of meat and meat products [12]. Various substances such as vinegar, wine, yogurt, fruit juices, spices, salt, oils, phosphates (alkaline, acidic), organic acids, and several compounds that give aroma are used in marinating [13, 14, 15].

In recent years, consumers have found that meat products prepared with natural additives are more reliable than conventional additives containing chemicals such as alkaline phosphatases [16]. Essential oils have a very important place among these additives. They have been used for thousands of years. Essential oils, which are colloquially called aromatic, ethereous or volatile oils, are mostly produced from various plants with aromatic properties in countries with tropical or temperate climates [17]. The antioxidant activity of aromatic plants and oils result from the phenolic compounds in their structures. These compounds contain mostly flavonoids, phenolic acids and phenolic terpenes [15, 18]. These substances are found in the leaves, flowers and woody parts of plants. Therefore, they are used in the form of medicine by drying the flowers and leaves of aromatic plants, or after obtaining essential oil by methods such as extraction and distillation [15, 19]. Many studies investigating the effects of essential oils used in food preservation have found that these oils have significant positive effects on food preservation [20, 21, 22]. Some herbal essential oils are accepted as GRAS (Generally Recognized as Safe) by the U.S. Food and Drug Administration (FDA) and they are classified as taste, odor and food additives. In the EU, volatile oils are used as safe food additives at the concentrations of less than 2 $mg \cdot kg^{-1}$ $bw \cdot day^{-1}$ [23]. Essential oils are known to have a potential effect on food preservation [17]. Studies have demonstrated that volatile oils of rosemary *(Rosmarinus officinalis)*, thyme (*Thymus vulgaris*) and clove (*Syzygium aromaticum*) have positive effects on the shelf life of chicken meat and products [3, 24, 25, 26].

This study used essential oils of rosemary with active substance of cineole, thyme with active substance of thymol-carvacrol and clove with active substance of eugenol. The purpose of this study was to investigate the efficacy of rosemary, thyme and clove essential oils added at different doses (125 $mg \cdot kg^{-1}$ and 250 $mg \cdot kg^{-1}$) on some microbiological, physicochemical and sensory properties of chicken breasts.

MATERIALS AND METHODS

Collection and preparation of samples

Fresh chicken breasts purchased in their original packaging from the local market in Elazig/Türkiye, regardless of the company name, were brought to the laboratory in the cold chain. They were diced in approximately 10 ± 0.1 g using a sterile knife and a sterile chopping board. They were placed into sterile bags and stored in the refrigerator at 4 ± 1°C until analysis (at 1 hour (h) and 24 h of marination. Then, the marination formulated below was prepared and experimental groups were formed.

Preparation of marination

Marinade ingredients were supplied from a local market. Tomato paste (200 g) (Tat Salça, Türkiye), sweet red pepper paste (200 g) (Tat Salça, Türkiye) (Bağdat, Türkiye), sunflower oil (250 mL) (Komili, Türkiye), freshly-squeezed lemon juice (200 mL), garlic (70 g), salt (45 g) (Billur Tuz, Türkiye) and spices [black pepper (10 g), cumin (10 g), red pepper flakes (15 g)] (Bağdat Baharat, Türkiye) were used in homemade marinade for chicken breasts. The marination sauce was homogenized with the help of a sterile blender (Prokit 444, Arzum, Türkiye) in a sterile container.

Adjusting the amounts of essential oils

The amounts of essential oils to be added to the marination were prepared based on the doses recommended by the manufacturer. A pilot study was performed while planning this study. The maximum dose of 500 $mg \cdot kg^{-1}$, a half dose of 250 $mg \cdot kg^{-1}$ and a quarter dose of 125 $mg \cdot kg^{-1}$ recommended by the manufacturer were added to the marination, thoroughly blended with chicken breasts and hold for 1 h. Then, they were cooked in the oven (MF 2009, Arçelik, Türkiye) at 200°C until browned (approximately 35–40 min). After that, they were evaluated sensorially by a group of 10 panelists (from academics at the Department of Food Hygiene and Technology, a group of 10 educated panelists of the same age and gender scored them each time) in terms of color, odor and taste. The maximum dose of 500 $mg \cdot kg^{-1}$ was definitely not accepted. Groups containing doses of 250 $mg \cdot kg^{-1}$ and 125 $mg \cdot kg^{-1}$ were accepted. Based on the sensory evaluation results, the following groups were formed.

Preparation of experimental groups

Marination was prepared in sterile bags. Groups were formed by adding essential oils of rosemary, thyme, clove at the doses of 125 $mg \cdot kg^{-1}$ and 250 $mg \cdot kg^{-1}$. No essential oils were added to the control group. Essential oils of rosemary (*Rosmarinus officinalis*) (Kalsec

20–01), thyme *(Thymus vulgaris)* (Kalsec 35–02) and clove *(Syzygium aromaticum)* (Kalsec 06–01) were supplied from KALSEC (MICHIGAN, USA). Microbiological analyses, pH measurement and sensory analyses of chicken breasts were performed at 1 h and 24 h of marination at 4°C. Experimental groups were prepared in double series. The study was repeated twice at an interval of 15 days.

Microbiological analyses

Aseptically, 10 g of chicken breast was placed into the special sterile bag of the homogenizer (Bag Mixer® 400, Interscience, France). It was homogenized by adding 90 mL of 0.1% sterile peptone water on it. Thus, 10–1 (1/10) dilutions of the samples were prepared. By using the same dilution and diluent, the samples were adjusted to 10–9 decimal dilutions. Microbiological cultivations were performed by both the pour plate method and the spread plate method in double parallel by taking 1 mL (pour plate method) and 0.1 mL (spread plate method) from each decimal dilution of the samples. The petri dishes containing 30 to 300 colonies were counted after they were incubated at the appropriate temperatures and times [27, 28].

Plate Count Agar (PCA) (Merck 1.05463.0500, Darmstadt, Germany) was used for total mesophilic aerobic (TMA) count (24–48 h at 35 ± 1°C) and total psychrophilic aerobic (TPA) count (5–7 ± 1°C for 7–10 days) [29], Violet Red Bile (VRB) Agar (Biokar BK152HA, Beauvais, France) for coliform bacteria count (24 h at 37 ± 1°C) [30], Violet Red Bile Glucose (VRBG) Agar (Biokar BK011HA, Beauvais, France) for *Enterobacteriaceae* count (24 h at 37 ± 1°C) [31], Tryptone Bile X Glucuronide (TBX) Agar (Merck 1.16122.0500, Darmstadt, Germany) for *Escherichia coli* count (4 h at 30 ± 1°C and then 18 h at 44 ± 1°C) [32], Dichloran Rose Bengal Chloramphenicol Agar (DRBC) (Biokar BK198HA, Beauvais, France) for yeast–mold count (5 days at 25 ± 1°C) [33], Baird Parker Agar (Oxoid CM0275, UK) for *Staphylococcus–Micrococcus* count (48 h at 37 ± 1°C) [34]. Baird–Parker Agar Base (BPA) containing Egg Yolk–Tellurite Emulsion (Oxoid SR0054C, UK) was used for *Staphylococcus aureus* count (48 h at 35 ± 1°C). Coagulase (+) *Staphylococcus aureus* count was determined by the coagulase test applied to gray–black, shiny colonies surrounded by clear zones formed in BPA medium [35].

Physicochemical analysis (pH)

The pH values of chicken breast samples (25 ± 1°C) were measured using a digital pH meter (HI 11310, Hanna Instruments, USA) [36]. For pH measurement, a 10 g sample was homogenized with 90 mL distilled water, and the measurement was made.

Sensory analyses

At both 1 h and 24 h, chicken breasts were cooked in an oven (MF 2009, Arcelik, Türkiye) at 200°C for approximately 35–40 min until they were completely browned. Then, using the sensory analysis form, a group of 10 educated panelists of the same age and gender scored them each time. Sensory evaluation was based on the panelists' basic smell and vision test. For marinated chicken breasts for which sensory examinations were performed at both 1 h and 24 h, their color, appearance, odor, crispness, taste and overall acceptability criteria were evaluated. For the sensory evaluation, the panelists drank water before each sample and evaluated the samples randomly [7, 37].

Statistical analyses

Descriptive statistics of data on microbiological, pH and sensory properties of marinated chicken breasts added with essential oils and the relationships between values were obtained using SPSS 21.0 (IBM SPSS, IBM Corporation, USA) package program. Microbiological data were calculated logarithmically and expressed as $\log_{10}$CFU·g^{-1}. Prior to analysis, a normality test using the Kolmogorov–Smirnov test and a homogeneity test using the Levene test were applied to all collected data. The One–Way ANOVA was used to compare the groups. Significance levels were determined by Duncan's test. The Independent T–Test was used to compare the sampling times. Values were given as mean ± standard deviation. Statistical significance level was accepted as $P \leq 0.05$ [38].

RESULTS AND DISCUSSIONS

Microbiological analysis findings of raw chicken breast are shown in TABLE I, microbiological analysis findings of marinated chicken breast in TABLE II, pH values of raw chicken breast and marinade sauce in TABLE III, pH values of marinated chicken breast in TABLE IV and sensory analysis findings of marinated chicken breast in TABLE V.

Microbiological analysis results

The average values of raw chicken breasts used in the analysis was found to be 5.46 $\log_{10}$CFU·g^{-1} for total mesophilic aerobic (TMA) count, 3.23 $\log_{10}$CFU·g^{-1} for total psychrophilic aerobic count, 1.24 $\log_{10}$CFU·g^{-1} for coliform count, 2.17 $\log_{10}$CFU·g^{-1} for *Enterobacteriaceae* count, 1.43 $\log_{10}$CFU·g^{-1} for *Staphylococcus–Micrococcus* count and 1.12 $\log_{10}$CFU·g^{-1} for yeast–mold count. No *E. coli* and *Staph. aureus* bacteria were found (TABLE I).

TABLE I
Microbiological Analysis Findings of Raw Chicken Breast Meat ($\log_{10}$CFU·g^{-1})

Microorganism	Mean ± Standard Deviation
Total Mesophilic Aerobic	5.46 ± 0.29
Total Psychrophilic Aerobic	3.23 ± 0.31
Coliform	1.24 ± 0.20
Enterobacteriaceae	2.17 ± 0.13
Escherichia coli	<1.00 ± 0.00
Staphylococcus–Micrococcus	1.43 ± 0.06
Staphylococcus aureus	<1.00 ± 0.00
Yeast–Mold	1.12 ± 0.07

Total mesophilic aerobic (TMA) count was found to be 5.25 $\log_{10}$CFU·g^{-1} in the control group and between 5.54 and 5.84 $\log_{10}$CFU·g^{-1} in the essential oil groups at 1 h. No statistical difference was observed between the control group and other groups at 1 h of storage (P>0.05) (TABLE II). At 24 h of storage, the count increased in the control group (6.35 $\log_{10}$CFU·g^{-1}) but there were decreases in all other groups. The highest inhibition was found in B2 group (2.49 $\log_{10}$CFU·g^{-1}) in which rosemary was used at a dose of 250 mg·kg^{-1}. This was followed by Ke2 (2.20 $\log_{10}$CFU·g^{-1}) and Ka2 groups (2.15 $\log_{10}$CFU·g^{-1}), respectively. There was a statistically significant difference between 1st h and 24th h in the other groups except the control group (P>0.05) (TABLE II). The study results were consistent with those of many studies showing that thyme essential oil had an inhibitory effect on the total mesophilic aerobic bacteria count [3, 39, 40, 41, 42, 43] (2–3 $\log_{10}$CFU·g^{-1}; 1 $\log_{10}$CFU·g^{-1};

1–5 $log_{10}CFU·g^{-1}$; 2 $log_{10}CFU·g^{-1}$; 2.3–3.1 $log_{10}CFU·g^{-1}$; 1.14 $log_{10}CFU·g^{-1}$, respectively). The result of clove essential oil (Ke2: 2.20 $log_{10}CFU·g^{-1}$) was similar to Fernández-Pan *et al.* [41] result (2 $log_{10}CFU·g^{-1}$), with that of rosemary essential oil (B1: 1.15 $log_{10}CFU·g^{-1}$) to Mahrour *et al.* [44] result (1.6 $log_{10}CFU·g^{-1}$).

Total psychrophilic aerobic bacteria are the predominant bacteria in chilled chicken meat and products. Knowing the microbiological load of the products is considered an indicator for the preservation or degradation of the quality of the products. [5, 45] Total psychrophilic aerobic (TPA) bacteria count was determined as 3.23 $log_{10}CFU·g^{-1}$ in raw chicken breasts (TABLE I). It was found to be 4.10 $log_{10}CFU·g^{-1}$ in the control group and 3.60–3.93 $log_{10}CFU·g^{-1}$ in the marinated groups added with essential oils at 1 h. At 24 h, the count of the control group increased to 5.40 $log_{10}CFU·g^{-1}$ but the counts of the marinated groups decreased to 1.20–2.90 $log_{10}CFU·g^{-1}$ (TABLE II). The highest bacterial inhibition was observed in Be2 (2.40 $log_{10}CFU·g^{-1}$), Ke2 (2.08 $log_{10}CFU·g^{-1}$) and Ka2 (2.04 $log_{10}CFU·g^{-1}$) groups, respectively. It was observed to be statistically significant differences between 1st h and 24th h between the control group and the marinated groups ($P<0.05$) (TABLE II). In addition, the bacterial inhibition level observed in B1 (1.13 $log_{10}CFU·g^{-1}$), Ke1 (1.01 $log_{10}CFU·g^{-1}$) and Ka1 (1.03 $log_{10}CFU·g^{-1}$) groups was similar to the 1 $log_{10}CFU·g^{-1}$ inhibition level detected by Fernández-Pan *et al.* study [41]. Also, the results obtained during the storage period in all the marinated groups were similar to the findings of some researchers [3, 46] stating that this group of bacteria decreased during the storage period.

TABLE II
Microbiological Analysis Findings of Marinated Chicken Breast Meat ($log_{10}CFU·g^{-1}$)

Microorganism	Analysis H	Groups						
		Control	B1	B2	Ke1	Ke2	Ka1	Ka2
TMA	1 h	5.25±0.31	5.61±0.04	5.54±0.06	5.71±0.03	5.62±0.11	5.84±0.05	5.78±0.07
	24 h	6.35±0.31^{a}	4.46±0.02^{b}	3.05±0.07^{d}	4.56±0.03^{b}	3.42±0.02^{dc}	4.39±0.41^{b}	3.63±0.14^{c}
TPA	1 h	4.10±0.04^{a}	3.68±0.02^{c}	3.60±0.25^{b}	3.81±0.01^{b}	3.73±0.07^{b}	3.93±0.02^{ab}	3.88±0.02^{ab}
	24 h	5.40±0.04^{a}	2.55±0.07^{d}	1.20±0.01^{f}	2.80±0.01^{c}	1.65±0.07^{f}	2.90±0.01^{b}	1.84±0.01^{e}
Coliform	1 h	2.02±0.08^{ab}	1.80±0.10^{bc}	1.57±0.09^{c}	2.15±0.21^{ab}	2.00±0.28^{ab}	2.38±0.11^{a}	2.20±0.14^{ab}
	24 h	1.05±0.07^{a}	<1.0^{b}	<1.0^{b}	1.10±0.14^{a}	<1.0^{b}	1.15±0.21^{a}	<1.0^{b}
Enterobacteriaceae	1 h	2.39±0.12^{bc}	2.29±0.04^{cd}	2.11±0.07^{d}	2.51±0.10^{ab}	2.28±0.02^{cd}	2.67±0.03^{a}	2.45±0.07^{bc}
	24 h	1.97±0.03^{a}	1.22±0.03^{c}	<1.0^{d}	1.45±0.01^{b}	<1.0^{d}	1.65±0.21^{b}	<1.0^{d}
Escherichia coli	1 h	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	24 h	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
Staphylococcus–Micrococcus	1 h	1.78±0.22^{a}	<1.0^{b}	<1.0^{b}	<1.0^{b}	<1.0^{b}	<1.0^{b}	<1.0^{b}
	24 h	1.10±0.14^{a}	<1.0^{b}	<1.0^{b}	<1.0^{b}	<1.0^{b}	<1.0^{b}	<1.0^{b}
Staphylococcus aureus	1 h	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
	24 h	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
Yeast–Mold	1 h	2.48±0.14^{a}	2.10±0.13^{bc}	1.90±0.14^{c}	2.30±0.12^{ab}	2.10±0.14^{bc}	2.42±0.16^{ab}	2.41±0.09^{ab}
	24 h	1.85±0.07^{a}	<1.0^{c}	<1.0^{c}	1.00±0.01^{b}	<1.0^{c}	1.00±0.01^{b}	<1.0^{c}

$^{a-f}$: Those with superscripts different from the averages in the same row are statistically significant ($P<0.05$); TMA: Total Aerobic Mesophilic ; TPA: Total Psychrophilic Aerobic; Control: Marinated chicken breast; B1: Marinated chicken breast added with 125 mg·kg^{-1} of rosemary essential oil; B2: Marinated chicken breast added with 250 mg·kg^{-1} of rosemary essential oil; Ke1: Marinated chicken breast added with 125 mg·kg^{-1} of thyme essential oil; Ke2: Marinated chicken breast added with 250 mg·kg^{-1} of thyme essential oil; Ka1: Marinated chicken breast added with 125 mg·kg^{-1} of clove essential oil; Ka2: Marinated chicken breast added with 250 mg·kg^{-1} of clove essential oil

Enterobacteriaceae bacteria are a group of bacteria that constitute an important part of the chicken meat microbiota [47]. *Enterobacteriaceae* count was found to be 2.17 $log_{10}CFU·g^{-1}$ in raw chicken breast (TABLE I). For the control group, it was detected to be 2.39 $log_{10}CFU·g^{-1}$ at 1 h and 1.97 $log_{10}CFU·g^{-1}$ at 24 h. A decrease was observed in *Enterobacteriaceae* count in all groups. The counts were below the limit of detection in Be2, Ke2 and Ka2 groups at 24 h. The highest inhibition was observed in Ka2 (1.45 $log_{10}CFU·g^{-1}$) and Ke2 (1.28 $log_{10}CFU·g^{-1}$) groups. These results were similar to those of the study demonstrating that thyme and clove essential oils led to a 1 $log_{10}CFU·g^{-1}$ decrease in *Enterobacteriaceae* bacteria count [41]. It was observed to be a statistically significant difference between 1st h and 24th h in all groups ($P<0.05$) (TABLE II). The relevant results were consistent with the findings of some researchers [3, 42, 43, 47, 48, 49].

Coliforms are one of the most common hygiene indicators found in foods. Also, their presence in foods is an indicator of both environmental and fecal contamination [1, 50]. This study showed the coliform count to be 1.24 $log_{10}CFU·g^{-1}$ in raw chicken breast (TABLE I). Coliform counts were found to be 2.02 $log_{10}CFU·g^{-1}$ in the control group and 1.57–2.38 $log_{10}CFU·g^{-1}$ in the marinated groups at 1 h of marination. At 24 h of marination, it decreased in all groups and was below the limit of detection (<1.0 $log_{10}CFU·g^{-1}$) in B1, B2, Ke2 and Ka2 groups. The highest inhibition was found in Ka2 (1.20 $log_{10}CFU·g^{-1}$) group. The difference was statistically significant in the coliform count in

all groups at 1 h and 24 h (P<0.05)(TABLE II). It was observed that the results obtained were consistent with those of some studies showing that the addition of thyme essential oil to chicken meat and products led to a decrease in the coliform counts [42, 46].

Escherichia coli bacteria count was below the limit of detection (less than 1.0 $\log_{10}$CFU·g^{-1}) in raw chicken breast (TABLE I). Therefore, no growth was observed in any of the marinated groups at both 1 h and 24 h (TABLE II).

Staphylococcus–Micrococcus count was found to be 1.43 $\log_{10}$CFU·g^{-1} in raw chicken breast (TABLE I). For the control group, it was detected to be 1.78 $\log_{10}$CFU·g^{-1} at 1 h and decreased to 1.10 $\log_{10}$CFU·g^{-1} at 24 h. However, this decrease was not found to be statistically significant (P>0.05) (TABLE II). *Staphylococcus–Micrococcus* count was below the limit of detection (<1.0 $\log_{10}$CFU·g^{-1}) in all the marinated groups at both 1 h and 24 h. The study results were consistent with the findings of some researchers stating that the essential oils used in chicken breasts had inhibitory effects on *Staphylococcus–Micrococcus* bacteria [51, 52]. *S. aureus* bacteria count was below the limit of detection (<1.0 $\log_{10}$CFU·g^{-1}) in raw chicken breasts (TABLE I) and in all the marinated groups (TABLE II).

Yeast–mold count is one of the species considered as an indicator of spoilage in poultry meats [4]. Yeast–mold count was 1.12 $\log_{10}$CFU·g^{-1} in raw chicken breast (TABLE I). For the control group, the yeast–mold count was found to be 2.48 $\log_{10}$CFU·g^{-1} at 1 h and 1.85 $\log_{10}$CFU·g^{-1} at 24 h. There were decreases observed in all groups during the storage period. Yeast–mold counts were below the limit of detection (<1.0 $\log_{10}$CFU·g^{-1}) in B1, B2, Ke2 and Ka2 groups at 24 h. The differences were observed to be statistically significant between the control group and all the marinated groups (P<0.05) (TABLE II). The highest bacterial count was found in B2 (1.90 log) group. It was similar to the findings of some researchers demonstrating that essential oils decreased yeast–mold counts in chicken breasts [3, 46, 47, 48, 53].

pH analysis results

The pH values of raw chicken breast and marinade sauce were found to be 5.50 and 3.85, respectively (TABLE III and FIGURE I). pH values were measured as 4.86 in the control group and 4.81–4.92 in the marinated groups at 1 h. These values increased partially at 24 h in all groups including the control group. These increases were statistically significant in the Control, Ke2, Ka1 and Ka2 groups (P<0.05)(TABLE IV). However, the increases in B1, B2 and Ke1 groups were not observed to be significant (P>0.05)(TABLE IV). The obtained results were similar to the findings demonstrating that the pH value increased during the storage period in Al–Hijazeen *et al.* study [43] on chicken breasts added with essential oils.

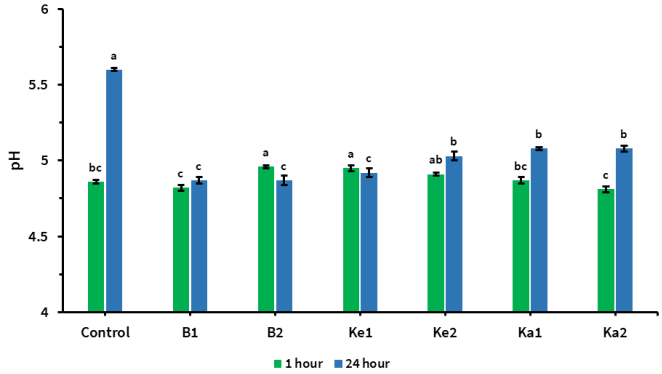

FIGURE 1. pH values of marinated chicken breast meat

Sensory analysis results

TABLE V shows the changes in the sensory properties (color, appearance, odor, crispness, taste, overall acceptability) of marinated chicken breasts at 1 h and 24 h. Accordingly, there was no significant change in the color and appearance scores of all groups, including the control group, at both 1 h and 24 h (P>0.05). Based on odor scores, a decrease was observed in the control group but an increase in B1, Ke1 and Ka1 groups and no change in B2, Ke2 and Ka2 groups at 24 h.

The differences were not significant between the control group and B1 and Ka1 groups in terms of odor scores (P>0.05). However, there were significant differences between the control group and B2, Ke1, Ke2 and Ka2 groups (P<0.05). Based on crispness and taste scores, an increase was observed in the control, B1, Ke1 and Ka1 groups but no change in Be2, Ke2 and Ka2 groups at 24 h. In terms of crispness scores, the differences were found to be significant between the control group and other groups (P<0.05) but not to be significant between the control group and Ka1 group (P>0.05). In terms of taste scores, there were no significant differences between the control group and B2, Ke2 and Ka1 groups (P>0.05). Based on overall

TABLE III
pH Values of Raw Chicken Breast Meat and Marinade Sauce

Analysis	Raw Chicken Breast Meat	Sauce
pH	5.50±0.02	3.85±0.07

TABLE IV
pH Values of Marinated Chicken Breast Meat

Analysis	Analysis H	Groups						
		Control	B1	B2	Ke1	Ke2	Ka1	Ka2
pH	1 h	4.86±0.01^{a}	4.82±0.02^{a}	4.80±0.01^{a}	4.92±0.02^{b}	4.91±0.01^{b}	4.87±0.02^{a}	4.81±0.02^{a}
	24 h	5.60±0.01^{c}	4.87±0.02^{a}	4.87±0.03^{a}	4.95±0.03^{b}	5.03±0.03^{c}	5.08±0.01^{c}	5.08±0.02^{c}

$^{a-c}$: Those with superscripts different from the averages in the same row are statistically significant (P<0.05); Control: Marinated chicken breast; B1: Marinated chicken breast added with 125 mg·kg^{-1} of rosemary essential oil; B2: Marinated chicken breast added with 250 mg·kg^{-1} of rosemary essential oil; Ke1: Marinated chicken breast added with 125 mg·kg^{-1} of thyme essential oil; Ke2: Marinated chicken breast added with 250 mg·kg^{-1} of thyme essential oil; Ka1: Marinated chicken breast added with 125 mg·kg^{-1} of clove essential oil; Ka2: Marinated chicken breast added with 250 mg·kg^{-1} of clove essential oil

TABLE V
Sensory Analysis Findings of Marinated Chicken Breast Meat

Analysis	Analysis H	Groups						
		Control	B1	B2	Ke1	Ke2	Ka1	Ka2
Color	1 h	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00
	24 h	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00
Appearance	1 h	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00
	24 h	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00	5.00±0.00
Odor	1 h	4.00±0.00^{a}	2.00±0.00^{c}	2.00±0.00^{c}	3.00±0.00^{b}	2.00±0.00^{c}	2.00±0.00^{c}	1.00±0.00^{d}
	24 h	3.00±0.00^{b}	3.00±0.00^{b}	2.00±0.00^{c}	4.00±0.00^{a}	2.00±0.00^{c}	3.00±0.00^{b}	1.00±0.00^{d}
Crispness	1 h	2.00±0.00^{b}	3.00±0.00^{a}	2.00±0.00^{b}	3.00±0.00^{a}	2.00±0.00^{b}	2.5±0.00^{ab}	1.00±0.00^{c}
	24 h	3.00±0.00^{c}	4.00±0.00^{ab}	2.00±0.00^{d}	4.50±0.70^{a}	2.00±0.00^{d}	3.50±0.70^{bc}	1.00±0.00^{e}
Taste	1 h	2.00±0.00^{b}	3.00±0.00^{a}	2.00±0.00^{b}	3.00±0.00^{a}	2.00±0.00^{b}	2.00±0.00^{b}	1.00±0.00^{c}
	24 h	2.50±0.70^{cd}	4.00±0.00^{b}	2.00±0.00^{d}	5.00±0.00^{a}	2.00±0.00^{d}	3.00±0.00^{c}	1.00±0.00^{e}
Overall Acceptability	1 h	42.50±3.53^{e}	57.5±3.48^{b}	52.50±3.50^{bc}	70.00±0.00^{a}	50.00±0.00^{d}	57.50±3.56^{b}	45.00±0.00^{de}
	24 h	52.50±2.48^{d}	69.00±1.41^{ab}	47.50±3.50^{d}	74.0±01.38^{a}	52.50±2.53^{d}	62.50±3.20^{c}	40.00±0.00^{e}
Total	1 h	60.50±3.46^{ef}	75.50±3.42^{b}	68.5±3.46^{cd}	89.00±0.00^{a}	66.00±0.00^{de}	74.0±04.24^{bc}	58.00±0.00^{ef}
	24 h	71.00±4.24^{c}	88.50±3.53^{b}	63.00±2.82^{d}	97.50±2.12^{a}	68.50±3.51^{cd}	82.00±4.24^{b}	53.00±0.00^{e}

$^{a–f}$: Those with superscripts different from the averages in the same row are statistically significant (P<0.05); Control: Marinated chicken breast; B1: Marinated chicken breast added with 125 mg·kg^{-1} of rosemary essential oil; B2: Marinated chicken breast added with 250 mg·kg^{-1} of rosemary essential oil; Ke1: Marinated chicken breast added with 125 mg·kg^{-1} of thyme essential oil; Ke2: Marinated chicken breast added with 250 mg·kg^{-1} of thyme essential oil; Ka1: Marinated chicken breast added with 125 mg·kg^{-1} of clove essential oil; Ka2: Marinated chicken breast added with 250 mg·kg^{-1} of clove essential oil

acceptability and total scores, a decrease was observed in B2 and Ka2 groups but an increase in other groups at 24 h. In terms of overall acceptability scores, there were differences between other groups (P<0.05) but no differences between the control group and B2 and Ke2 groups (P>0.05). The differences were significant between the control group and all other groups in terms of total score (P<0.05). Based on scoring, the most acceptable group was Ke1 group with 97.50 points. This was followed by B1 (88.50), Ka1 (82.00) and control group (71.00), respectively. Ka2 (53.00) group had the lowest score. Ke1 group was accepted, which was consistent with the results of some researchers stating that thyme essential oil was accepted in chicken breasts [3, 47, 54].

It was found that 250 mg·kg^{-1} doses of essential oils, especially at 24 h, and rosemary had more inhibitory effects on some microbial (TMA, TPA and yeast–mold) parameters. However, the sensory groups with the addition of 125 mg·kg^{-1} were more accepted. Among these groups, the most acceptable group was the group added with 125 mg·kg^{-1} of thyme essential oil.

CONCLUSION

The use of essential oils in chicken breast can be recommended as a natural preservative. Microbiological analyses have demonstrated that marinated chicken breasts added with different doses of essential oils could control the growth of microorganisms at 4°C and at 1 h and 24 h of storage. In particular, it was concluded that the use of thyme essential oil as an alternative to chemical preservatives may be beneficial both in extending the shelf life of marinated chicken breasts and in terms of consumer taste. This study presents a wide range of use of natural preservatives for healthier poultry meat. Due to short shelf life and quality defects, the use of mixtures containing essential oils in meats will also provide significant benefits to public health. Thus, this study strongly supports the claim that natural preservatives can be replaced by chemical preservatives in ready-to-cook poultry meats without reducing quality, shelf life and consumer acceptance.

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Ethics statement

This study was conducted in Elazig with the approval of Firat University Non-Interventional Research Ethics Committee with protocol number 2023/04-25 and dated March 09, 2023.

Conflict of interest statement

The authors declare that they have no conflict of interest

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