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Application of essential oils as antibacterial substances in minced meat in Al-Najaf city, Iraq

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ABSTRACT

Background: Meat has been regarded as a very healthy food because of its nutritional value; minced meat is a popular meal; however, microbial contamination may have negative impacts on meat quality and consumer health.
Aim: The study aimed to estimate the bacteriological contamination and use of some chemical and natural preservatives in minced meat.
Methods: Approximately 103 samples of minced meat were collected from different butchers in Al-Najaf province from December 2024 to March 2025 to monitor their microbiological quality. Biochemical and polymerase chain reaction techniques for 16S rRNA gene amplification were used to confirm the bacterial identity, and the effects of lactic acid (3%), rosemary volatile oil (2%), thyme oil (1.5%), and the combination of thyme (1.5%) and rosemary (2%) were used as antibacterial after immersing the samples with these substances following bacterial counts.
Results: Total coliforms, *Escherichia coli,* and *Staphylococcus aureus* were 70%, 58%, and 54%, respectively. Furthermore, the mean values of average bacterial counts of total coliform, *E. coli,* and *S. aureus* were 4.35, 3.73, and 3.43 CFU log10/g, respectively. Compared with the control, statistically significant differences ($p < 0.05$) were observed between the single and combination use of substances and lactic acid. The combination of rosemary and thyme was the most effective, resulting in a reduction of coliform, *E. coli*, and *S. aureus* (1.11, 1.95, and 1.42 CFU, respectively) after 7 days.
Conclusion: The high levels of contamination detected in the minced meat are an indication of unhygienic production conditions, and the synergistic activity of rosemary and thyme was most effective in reducing contamination.
Keywords: Contamination, *E. coli*, Essential oil, Minced meat, *S. aureus*.

Introduction

Meat is an essential component of the human diet because of its composition. However, its high moisture content, abundance of nitrogenous substances (including proteins and essential amino acids), and availability of minerals, vitamins, and other growth factors make meat and its products a perfect environment for bacterial growth (Hwang *et al.,* 2020). Furthermore, its pH level encourages microbial proliferation (Alahakoon *et al.,* 2015). Minced meat is a popular food frequently used as an ingredient in numerous dishes. After an animal is slaughtered, the meat is prepared into retail or wholesale cuts. The meat is then further processed and ground, often including trim and other cuts. This process significantly expands the surface area of the meat, which in turn promotes bacterial development and enhances their adhesion to the meat (Donsí *et al.*, 2011). High loads of microorganisms, especially pathogens, severely impact the meat's sanitary quality. These pathogens can contaminate food when handled in unhygienic conditions (Berhanu *et al.*, 2025). These microorganisms may spread through external exposure, a lack of hygiene in the slaughterhouse, tools, clothing, and the operator hands (Bantawa *et al.,* 2018). Inappropriate handling, cooking, and storage of meat and meat products contribute to a chain of food-borne bacterial infections that also includes food consumers (Berhanu *et al.*, 2025). Therefore, detecting and identifying pathogenic bacteria in food as well as determining the overall microbial load of the food are extremely important (Sachindra *et al.*, 2005). Indicator bacteria, such as *Escherichia coli* and coliform, are commonly used to evaluate the hygienic condition of food and their potential for infection. Moreover, these bacteria are vital indicators of food safety (Kozačinski *et al.,* 2006). *Escherichia coli* is the most common bacterial contaminant and a reliable indicator of fecal

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contamination in milk, water, food, and other products (El-Gendi and Mansy, 2017). On the other hand, there are at least 23 species in the staphylococcal genus, the most important of which is *Staphylococcus aureus* and the meat and its industries are extremely concerned about this pathogen (Hannan *et al.*, 2008). Furthermore, the presence of coliform immediately after production is a sign of fecal contamination in the water used to prepare the meat and human contamination by handlers (Soepranianondo *et al.,* 2019).

Several antibacterial compounds have been applied to meat to prolong its shelf life and maintain its freshness (Alparslan *et al.,* 2019). The food industry is now using natural antimicrobial agents from plants, animals, and microbes to combat health issues such as allergies, intoxication, and cancer, which can result from the use of systemic antimicrobial preservation (Field *et al.*, 2015). Rosemary (*Rosmarinus officinalis*), a member of the Lamiaceae family, is one such plant that is used in food products for its antioxidant and antimicrobial properties (Azizkhani and Tooryan, 2015). Similarly, thyme (*Thymus vulgaris*) is another plant used for its high content of phenolic compounds, which act as both antimicrobials and antioxidants in meat products (Bensid *et al.*, 2014). Beyond plant-based solutions, the application of 1.5% lactic acid to red meat has been shown to effectively lower *E. coli* levels (De Martinez *et al.*, 2002). Microbial contamination of whole pieces of meat typically occurs on the exposed surface; however, these surface-active microorganisms become distributed throughout the entire product when meat is minced. Consequently, it is crucial to monitor meat and its products. Therefore, the current study aims to assess the microbiological quality of minced meat and investigate the possibility of reducing the microbial load using natural antimicrobial preservatives in Al-Najaf city.

Materials and Methods

Study duration and location

This study was conducted in Al-Najaf city from December 2024 to March 2025. All samples were randomly obtained from city's butcher shops. The samples were sent in a cold box to the University of Kufa/Faculty of Veterinary Medicine/Laboratory of Public Health Department for further analysis.

Sample preparation for microbiological analysis

Under aseptic conditions, 103 samples of minced meat were collected (250 grams for each). The samples were homogenized and 10 grams from 250 grams were transferred to a sterile stomacher bag sack containing 90 ml of sterile 0.5% peptone water (Himedia). Using 10-fold serial dilutions, samples were serially diluted up to 10^{-7} (Erdem *et al.*, 2014).

Essential oil (EO) preparation

The EOs of rosemary (*Rosemarian officinalis*) and thyme (*Thymus vulgaris*) were extracted using hydrodistillation. Approximately 100 g of each pulverized plant material was added to a 2-l flask containing 1.5 l of double-distilled water. The mixture was then boiled for 4 hours. To extract the EO, the resulting vapor was condensed. The extracted oil was subsequently dried using anhydrous sodium sulfate. All EO was stored at freezing temperatures until its utilization (Abozid and Asker, 2013).

Bacterial counts

Empty sterile petridish were filled with 1 ml of the tested sample from each dilution (10^{-1} to 10^{-7}), which had been previously made from minced meat. Then, 10 to 15 ml of sterile VRB agar (Himedia) was added, after which a covering layer of approximately 3 ml of medium was poured as overly (John *et al.*, 2001). Plates were incubated at 37°C for 24–48 hours to check for total coliform. On VRB agar, dark red colonies were normal coliforms (Mansour *et al.,* 2019). According to Difco-Manual (1984), 0.1 ml of each duplicated dilution was spread on the eosin methylene blue agar (Himedia) using a sterile L-shaped bent glass. The plates were then allowed to dry, inverted, and incubated for 24–48 hours at 37°C. Colonies with a metallic green sheen were positive. Presumptive coagulase-positive staphylococci were isolated from the minced meat using mannitol salt agar and Baird parker agar (Himedia) (Pinamonti *et al.*, 2025). Using a sterile bent glass spreader, 0.1 ml from each of the previously made serial dilutions was applied to duplicate plates of Mannitol salt and Baird parker agars followed by incubation at 37°C for 48 hours. The total staphylococci count per gram was estimated after counting the white, orange, and yellow colonies on mannitol and the black colonies on Baird parker agar (Sadeghian *et al.,* 2025). Biochemical tests were performed on each positive isolate of *S. aureus* and *E. coli*, including catalase, coagulase, Latex mast staph, oxidase, and indol production. *Staphylococcu aureus* produced positive results for coagulase, catalase, and latex mast staph. *Escherichia coli* and coliform produced positive results for catalase because they all produced catalase enzyme, and produced positive results for oxidase and indol production.

Bacterial identification

DNA extraction

Genomic DNA was extracted from bacteria using the G-spin DNA extraction kit. A polymerase chain reaction (PCR) experiment using a specific primer was conducted by integrated DNA technologies (Integrated DNA Technologies company/USA) (Table 1).

25 µl of PCR amplification mixture containing (5 µl) Taq PCR premix, (1 µl) forward primer, (1 µl) reverse primer, (1.5 µl) DNA, and (16.5 µl) distilled water. 5 minutes for initial denaturation at 94°C was the PCR condition protocol. Denaturation-2 94°C for 45 seconds, annealing 56°C for 45 seconds, extention-1 72°C for 45 seconds, and extention-2 72°C for 7 minutes.

Table 1. The specific primer 16SRNA of gene.

Primer	Sequence	T_m (°C)	GC (%)	Product size
Forward	5′- AGAGTTTGATCCTGGCTCAG - 3′	54.3	50.0	1,250 base pair
Reverse	5′- GGTTACCTTGTTACGACTT - 3′	49.4	42.1	

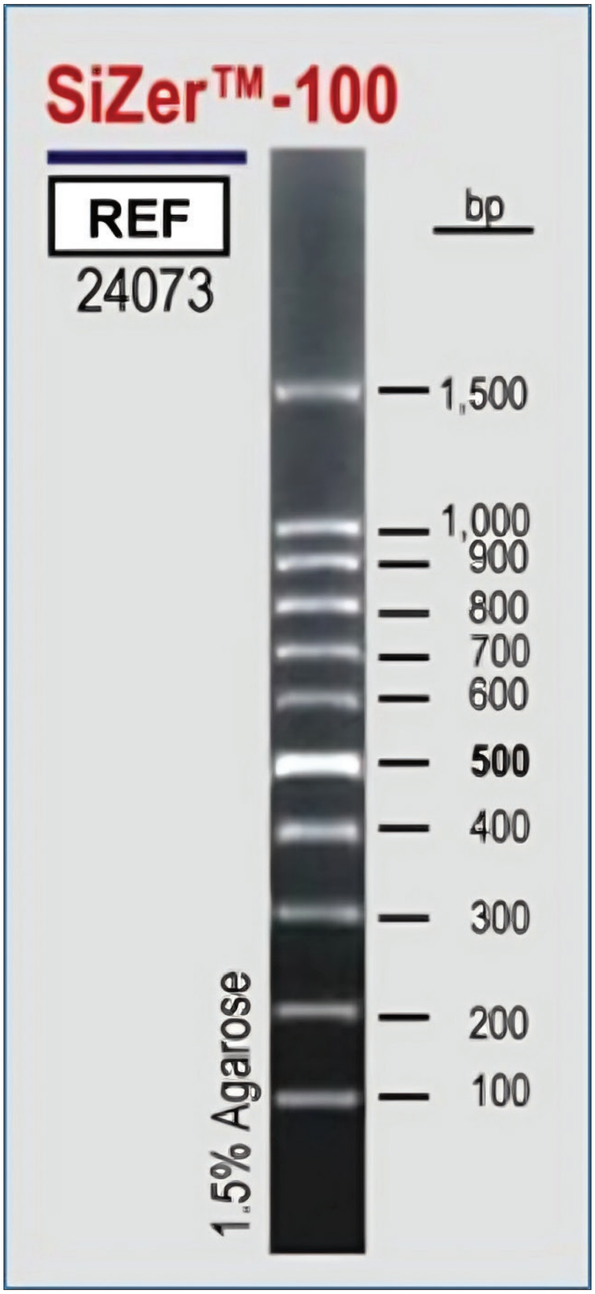

Fig. 1. 16SrRNA gene (15,00 bp) amplified PCR products: Agarose gel electrophoresis, 1.5% agarose, ethidium bromide staining, and 1.30 hours of electrophoresis at 5 volt/cm[2].

The 16S rRNA gene was amplified using the PCR technique to confirm the identification of bacterial species level, and the positive result was detected by 1.5% agarose gel electrophoresis, as shown in (Figs. 1-3).

Decontamination process

In this study, each sample was divided into five groups after bacterial counts (50g for each) to evaluate the preservatives used as decontamination. A1 as (untreated control group) without any preservative, A2 (positive group) was immersed in 2% rosemary oil (*Rosemarian officinalis*) for 10 minutes (Abed *et al.,* 2021), A3 was immersed in 1.5% thyme oil (*Thymus vulgaris*) for 30 minutes (Salán *et al.,* 2006), A4 was immersed in 3% lactic acid for 10 minutes (Khalafalla *et al.*, 2016), and A5 was immersed in a combination of thyme oil 1.5 and rosemary oil 2% for 10 minutes. All groups were stored at 4°C and examined for bacteriological examination after 3 and 7 days.

Statistical data analysis

The data were statistically analyzed using Statistical Analysis System (SAS; version 9.1). The *t* test was used to assess significant differences between the means of the two groups (SAS, 2010).

Ethical approval

No animal-related or animal-handling topics were included in this study. As a result, there was no ethical approval committee requirement.

Results

Table 2 shows the variation in the percentages of total coliform, *E. coli* and *S. aureus* isolated from minced meat purchased from butcher shops in Al-Najaf city. The results found that there were significant differences ($p < 0.05$) among the minced meat samples; among 103 samples, 70% (71/103) were contaminated with total coliform. The incidence of *E. coli* bacteria was 58% (60/103) and *S. aureus* was 54% (56/103). The isolation percentage according to the months of total coliform, *E. coli* and *S. aureus* showed a significant variation in the minced meat. March showed the highest isolation percentages of all bacterial counts Table 2.

There were significant differences ($p < 0.05$) in the total coliform in the minced meat according to the months (Table 3). The total coliform was less in December and January than in February and March. The values of total coliform in minced meat in December and January were 3.65 and 4.33 CFU log_{10}/g, respectively, while the highest value was in March at 5.13 CFU log_{10}/g.

Escherichia coli counts with a mean value in the minced meat were 3.07 and 3.35 CFU log_{10}/g in December and January, respectively, which were less than in March, which was higher significantly ($p < 0.05$) at 4.97 CFU

Table 2. The isolation percentages % of total coliforms, *E. coli*, and *S. aureus* that were isolated from minced meat samples in different regions of Al-Najaf.

Months	Total coliform	*Escherichia coli*	*Staphylococcus aureus*
December	12/24 (50%)	10/24 (41%)	12/24 (50%)
January	17/24 (70%)	14/24 (58%)	12/24 (50%)
February	15/25 (60%)	12/25 (50%)	14/25 (56%)
March	27/30 (90%)	24/30 (80%)	18/30 (60%)
Total	71/103 (70%)	60/103 (58%)	56/103 (54%)
p-value	0.18	0.33	0.49

Table 3. Total coliforms, *E. coli*, and *S. aureus* counts (mean ± SE) (CFU log_{10}/g) in the minced meat samples collected from different regions of Al-Najaf according to the months.

Months	Total coliform	*Escherichia coli*	*Staphylococcus aureus*
December	3.65 ± 0.52^{c}	3.07 ± 0.49^{c}	2.32 ± 0.37^{c}
January	4.33 ± 0.97^{cb}	3.35 ± 0.38^{cb}	2.97 ± 0.26^{bc}
February	4.30 ± 0.87^{b}	3.52 ± 0.23^{b}	3.24 ± 0.28^{ab}
March	5.13 ± 0.22^{a}	4.97 ± 0.17^{a}	4.08 ± 0.18^{a}
Average	4.35 ± 0.65^{ab}	3.73 ± 0.32^{ab}	3.43 ± 0.24^{b}
LSD	0.92	0.53	0.41

Means in the same column with different small letters differ significantly ($p < 0.05$).

Table 4. Total coliform (Log_{10} CFU/g) decontamination results.

Group	A1`	A2	A3	A4	A5
Time			**3 days**		
Total coliform	4.06 ± 0.14^{a}	3.41 ± 0.31^{ab}	3.51 ± 0.26^{b}	3.21 ± 0.27^{c}	3.01 ± 0.26^{d}
Differences count	-	0.65	0.55	0.85	1.05
Time			**7 days**		
Total coliform	4.05 ± 0.20^{a}	3.05 ± 0.32^{ab}	3.15 ± 0.34^{b}	2.95 ± 0.32^{c}	2.75 ± 0.30^{d}
Differences count	-	1.01	0.91	1.11	1.31

Means in the same raw with different small letters differ significantly ($p < 0.05$).

log_{10}/g (Table 3). The minced meat was significantly more contaminated during March than other months ($p < 0.05$).

There were significant differences between the values of staphylococcus counts according to month (Table 3). The minced meat was significantly ($p < 0.05$) more contaminated in March (4.08 CFU log10/g) than in other months, while the lowest value was in December (2.32 CFU log10/g). The average value was 3.43 CFU log10/g.

Table 4 shows the decontamination effect of EOs (rosemary and thyme) and lactic acid on the inhibition of coliform. The current study found that lactic acid and the EO of rosemary and thyme affected coliform levels, but the combination of EO (A5) showed a static effect ($p < 0.05$) after 3 and 7 days.

Group A5 had the greatest decrease in coliform (1.05 and 1.31 Log_{10} CFU/g) after 3 and 7 days, respectively, compared with the control group.

Table 5 shows the decontamination effect of lactic acid and EO of rosemary and thyme on the inhibition of *E. coli*. The decrease in *E. coli* counts was significant ($p < 0.05$) in groups A2–A5 when compared to A1 (control group) after 3 and 7 days. The highest decrease was observed in lactic acid (A1) and the combination of rosemary and thyme (A5) (1.43 and 1.67 Log_{10} CFU/g) (1.95 and 1.84 Log_{10} CFU/g) after 3 and 7 days, respectively, but the greatest decrease was observed in lactic acid (A4) after 7 days (1.95 Log_{10} CFU/g) statistically ($p < 0.05$).

The change of *S. aureus* counts after using lactic acid and EO is shown in Table 6: All groups significantly exceeded the control group in reducing *S. aureus* after

Table 5. *E. coli* (Log_{10} CFU/g) decontamination results.

Groups	A1	A2	A3	A4	A5
Time			**3 days**		
E. coil counts	4.19 ± 0.19[a]	3.04 ± 0.23[ab]	3.06 ± 0.23[b]	2.76 ± 0.14[c]	2.52 ± 0.07[d]
Differences count	-	1.15	1.13	1.43	1.67
Time			**7 days**		
E. coli counts	4.15 ± 0.20[a]	2.95 ± 0.13[b]	2.89 ± 0.14[ab]	2.20 ± 0.10[d]	2.31 ± 0.03[c]
Differences count	-	1.2	1.26	1.95	1.84

Means in the same raw with different small letters differ significantly ($p < 0.05$).

Table 6. *Staphylococcus aureus* (Log_{10} CFU/g) decontamination results.

Groups	A1	A2	A3	A4	A5
Time			**3days**		
Staphylococcus aureus counts	2.94 ± 0.19[a]	2.45 ± 0.03 [ab]	2.51 ± 0.07[b]	2.16 ± 0.12[cb]	1.86 ± 0.15[d]
Differences count	-	0.49	0.43	0.78	1.08
Time			**7 days**		
Staphylococcus aureus counts	2.91 ± 0.20[a]	2.24 ± 0.22[b]	2.05 ± 0.24[ab]	1.49 ± 0.11[c]	1.30 ± 0.03[d]
Differences count	-	0.67	0.86	1.42	1.61

Means in the same raw with different small letters differ significantly ($p < 0.05$).

Achbele, E., Erku, W., Gebre-Michael, T. and Ashenafi, M. (2006). Cockroach-associated food-borne bacterial pathogens from some hospitals and restaurants in Addis Ababa, Ethiopia: distribution and antibiograms. J. Rural Trop. Public Health 5, 34–41.

3 and 7 days ($p < 0.05$). The greatest decrease in *S. aureus* counts was in the combination of rosemary and thyme EO (group A5) after 7 days (1.61 Log_{10} CFU/g) statistically ($p < 0.05$).

Discussion

Microbial contamination accounts for one-fourth of the global food supply loss, which has developed into a major ethical and economic issue globally (Huisin't 1998; Gavrilova *et al.*, 2019). Raw meat is one of the sources of foodborne infections in people because of its chemical components, which encourage the growth of a wide variety of bacterial populations (Doulgeraki *et al.,* 2012). In addition, meat may become contaminated during the slaughtering process by bacteria found in water, on floors, in utensils, in slaughter instruments, and through contact with people (Klaharn *et al.,* 2022). The percentage of total coliform, *E. coli,* and *S. aureus* showed significant variation according to the months, with the percentage being higher during March than other months. This might be because there are fewer refrigeration conditions and high ambient temperatures, which favor bacterial development and multiplication (Azage and Kibret, 2017). McEvoy *et al.* (2004) investigated the impact of the season on microbial contamination and found that the peak of bacterial contamination of carcasses increased with increasing ambient temperature. The isolation percentage disagreed with that reported by Ragab *et al.* (2016), who reported that the percentage of total coliforms was 100%, *E. coli* was 50%, *Staphylococcus* was 20%, and *Salmonella* was 20% of the total of 50 samples of minced meat in Egypt.

The coliforms activity was present in all of the samples from the study area. These bacteria are commonly found in water, food, and vegetable waste and cause infections of the bone or joint tissue as well as urinary tract infections in both humans and animals (Drzewiecka, 2016). The increase in coliform counts is consistent with an increase in microbial load, particularly pathogenic species, which causes an increase in coliform counts (Mercy *et al.*, 2022). Minced meat can become contaminated with coliform from a variety of sources, including an unhygienic cutting process; workers' hands; water used to clean meat or cutting tools; improperly cleaned meat grinding equipment (this was also observed as well during sample collection); and the meat itself because the coliform bacteria's natural habitat is the intestines of animals (Said *et al.*, 2021).

In recent years, *E. coli* has been regarded as the leading cause of food poisoning. Our results showed that the total percentage of *E. coli* from minced meat was 23/40 (57%) with an average value of 3.73 CFU log_{10}/g,

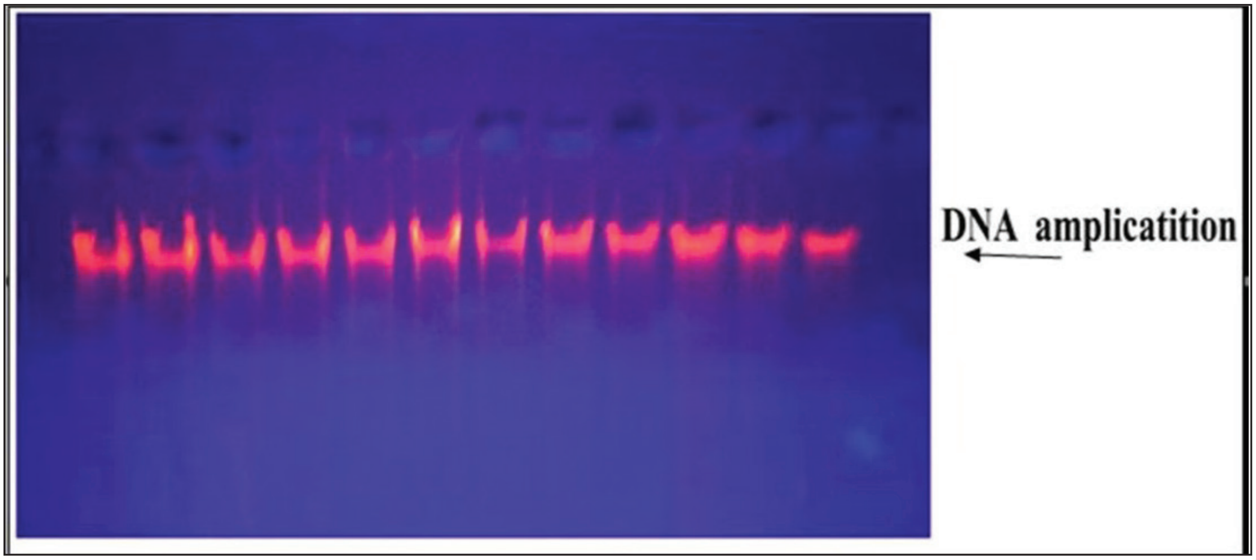

Fig. 2. Gel electrophoresis of genomic DNA extraction from bacteria, 1% agarose gel at 5 Vol/cm for 30 minutes.

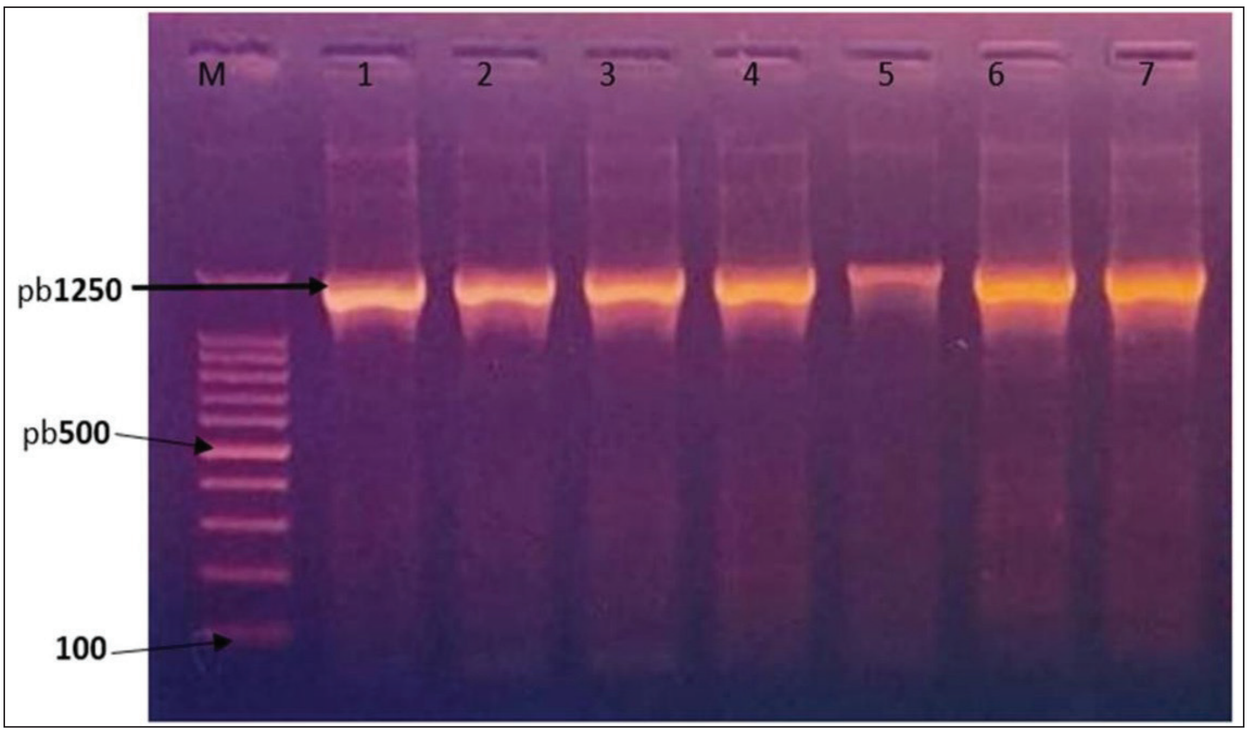

Fig. 3. PCR product the band size 1,250 bp. The product was electrophoresis on 1.5% agarose at 5 volt/cm^2. 1× TBE buffer for 1:30 hours. N: DNA ladder (100).

which agrees with Othman *et al.* (2023), who reported that the percentage of *E. coli* isolated from meat was 21/50 (42%). Our findings were also more significant than those of other research, which found that the prevalence rates of *E. coli* in meat were 1.5% in Iran (Siavash *et al.*, 2016), 17.8% in Australia, and 21.1% (49/232) in the United States (Othman *et al.*, 2023) and less than that reported by Siavash *et al.* (2016) who found 50% of *E. coli* in minced meat and the mean values counts was 6×10^4 in Egyptian retail markets. The difference in values between countries may be due to the high ambient temperature and the poor sanitary conditions in Iraq compared with those in other countries. Contamination of meat with *E. coli* may occur from the time animals reach the slaughterhouse until it is time to consume the meat. *Escherichia coli* is spread to meat and its products by a number of different methods, including animal skin, cutting equipment, unclean environments, and personnel who no longer practice good hygiene (Tate *et al.,* 2021).

Tables 2 and 3 show the percentage and enumeration of *S. aureus* isolates from minced meat in Al-Najaf city. The result in the current study was higher than that of Boukary *et al.* (2012)who found that the *S. aureus* in minced meat was 25% (2/8) and the mean value was 3.1 CFU/g, and less than that Ragab *et al.* (2016) who recorded that the *S. aureus* in minced meat was 20% and the mean average value was 3×10^3. The high value of *S. aureus* in March might be due to storage temperature and pH level that increased with increasing ambient temperature (Diyantoro and Wardhana, 2019). Meat plays a role as food for many bacteria, and these bacteria are considered a major source of meat contamination. Meat processing workers are one of the main causes of elevated staphylococcal counts on the surface of studied carcasses because their hands have been discovered to be highly contaminated; this is in agreement with the results of Ercolini *et al.* (2009), who reported that the main sources of contamination with Staphylococcal spp. during meat preparation are contaminated skin, feces, the contents of digestive organs, butcher’s knives, hands, clothes, and polluted water.

In the present study, the bacteriological quality of locally produced minced meat with coliform, *E. coli,* and *S. aureus* was investigated to examine the antimicrobial activity of lactic acid, rosemary, and thyme. The reduction percentage in samples immersed in thyme is similar to that obtained by Schlegelová *et al.* (2004), who found the reduction percentages to be 1.5%, 2%, and 2.5% when thyme oil was added to preserve minced meat. Recently, the antimicrobial activity of plant extracts has been studied in this research (Zaid *et al.*, 2021). The presence of carvacrol and thymol in the *Thymus vulgaris* and their ability to break down the outer membrane of gram-negative bacteria and increase the permeability of the cytoplasmic membrane have been reported that the EOs had excellent antimicrobial effects against *E. coli* and *S. aureus* (Boskovic *et al.*, 2015). Conversely, the reduction of *E. coli*, *S. aureus*, and coliform in the group that was immersed with rosemary (A2) was similar to the result found by Abed *et al.* (2021), who showed no CFU/g after 9 days of preservation when adding rosemary volatile oil to the canned meat sample. The antimicrobial activity of rosemary is attributed to its high levels of phenolic compounds, which have an antimicrobial effect against microorganism (Najem and Ibrahim, 2017). The reduction in counts of coliform, *E. coli*, and *S. aureus* in the samples immersed in lactic acid (A4) was similar to that reported by Najem and Ibrahim (2017). They reported that the lactic acid was most effective in decreasing the *E. coli* count to 1.92 CFU/g in the shrimp sample. In addition, our results agree with those Khalafalla *et al.* (2016), who found that the lactic acid had the greatest effect on the reduction of coliform in the chicken breasts.

Thyme and rosemary and their constituents, such as thymol and carvacrol are preservatives that could significantly affect the shelf life of meat (Boskovic *et al.,* 2015). Rosemary (*Rosmarinus officinalis*) and thyme (*Thymus vulgaris*) are safe food preservatives with strong antibacterial action against pathogenic bacteria such as *Salmonella enteritidis*, *Pseudomonas aeruginosa*, *Bacillus cereus, E. coli*, and *S. aureus* (Gonelimali *et al.,* 2018). The components in EOs, particularly phenolic compounds like carvacrol and thymol, have a unique hydrophobic nature that makes it easier for them to penetrate and divide within the lipid bilayer of the bacterial cell membrane. The structural integrity of the membrane is severely disrupted by this molecule interaction, which increases the membrane's permeability ().

The greatest reduction in coliform, *E. coli,* and *S. aureus* was observed in the combination of thyme and rosemary oil in group (A4). This may be due to the synergistic mechanism of active ingredients in the both thyme and rosemary EOs, such as eucalyptol, camphor, and thymol (Yalçın and Polat, 2022). Furthermore, the synergistic activity of EO may damage the cell wall by chemical substances (García-Díez *et al.,* 2017).

Conclusion

The minced meat was contaminated with more than acceptable levels of total coliform, *E. coli,* and *S. aureus* due to the meat being passed through different contamination stages, starting from the slaughterhouse until the sale of meat in the supermarket. This may be related to the lack of implementation of the inspection act and appropriate sanitary procedures in regard to meat, which suggests a possible risk of infection while consuming such food. The contamination was greater in March due to the high temperature and lack of refrigeration than in other months, and this may be because the minced meat is subjected to a lot of processing, starting with cutting the meat and putting it in the mincing machine, and the meat may be mixed with a lot of viscera and internal organs. Rosemary and thyme contain antimicrobial volatile oils that reduce contamination. This combination was more effective than using rosemary, thyme, or lactic acid alone.

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Conflict of interest

The authors declare that there is no conflict of interest.

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Author contribution

M.A.R: conceptualization, data collection, and writing of the original manuscript draft. A.A.S.: review and editing. All authors have approved the final manuscript for publication.

Data availability

All data supporting the findings of this study are available in the manuscript, and no additional data sources are required.

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